Plant responses to diversity-driven selection and associated rhizosphere microbial communities

Hahl, Terhi ; van Moorsel, Sofia J ; Schmid, Marc W ; Zuppinger-Dingley, Debra ; Schmid, Bernhard ; Wagg, Cameron

Abstract: Plant diversity loss can alter plant–plant and plant–rhizosphere microbiome interactions. These altered interactions, in turn, may exert diversity-driven selection pressure to which plants respond with phenotypic changes. Diverse plant communities may favour the survival and fitness of individuals with traits that avoid competition. Conversely, monocultures may accumulate species-specific pests favouring greater investment in defence traits. Yet, it is unknown how altered plant rhizosphere interactions influence the plant diversity-driven selection for altered plant phenotypes. We tested for plant diversity-driven selection on plant above-ground traits and how these traits are modified by their rhizosphere microbial communities after 11 years in experimental plant monocultures and mixtures. Plants propagated from monocultures or mixtures were grown in combination with their 'home' versus 'away' arbuscular mycorrhizal fungi (AMF) or non-AMF microbes in two separate experiments using five and eight plant species, respectively. We hypothesized that plants in monocultures may be selected for better defence and better performance in association with rhizosphere microbial communities compared with plants in mixtures. Monoculture and mixture plants significantly differed in their above-ground phenotypes. As predicted, plant traits related to defence (greater leaf mass per area and leaf dry matter content, reduced leaf damage) were more pronounced in monoculture plants in both experiments. Effects of the rhizosphere microbial communities, which generally enhanced plant growth, tended to be species-specific. Significant three-way interactions between diversity-driven selection, AMF treatment and plant species showed that home versus away effects could be positive or negative, depending on plant species. We conclude that long-term differences in plant diversity lead to selection for altered plant phenotypes. Such differences may be further modified in association with the AMF microbial communities derived from the different plant diversity treatments, but often outcomes are species-specific. This suggests that plant species differ in their capacity to respond to diversity loss and associated changes in rhizosphere microbial communities, making it complicated to predict community-level responses to such loss.

DOI: https://doi.org/10.1111/1365-2435.13511

DOI: https://doi.org/10.1111/1365-2435.13511

Plant responses to diversity-driven selection and associated rhizosphere microbial communities

Terhi Hahl[1]§, Sofia J. van Moorsel[1,2]§, Marc W. Schmid[1,3], Debra Zuppinger-Dingley[1], Bernhard Schmid[4] and Cameron Wagg[1,5]*

1. Department of Evolutionary Biology and Environmental Studies, University of Zürich, Winterthurerstrasse 190, CH-8057 Zurich, Switzerland
2. Department of Biology, McGill University, 1205 Penfield Ave., H3A 1B1, Quebec, Canada.
3. MWSchmid GmbH, Möhrlistrasse 25, CH-8006 Zurich, Switzerland
4. Department of Geography, University of Zürich, Winterthurerstrasse 190, CH-8057 Zurich, Switzerland
5. Fredericton Research and Development Centre, Agriculture and Agri-Food Canada, 850 Lincoln Road, P.O. Box 20280, Fredericton, E3B 4Z7 New Brunswick, Canada

§Equal contribution

*Author for correspondence:

Cameron Wagg, cameron.wagg@canada.ca

Abstract

1. Plant diversity loss can alter plant interactions and rhizosphere microbial communities. These altered interactions in turn exert diversity-driven selection pressures to which plants may respond with phenotypic changes. Diverse plant communities may favour the survival and fitness of individuals with traits that avoid competition. Conversely monocultures may accumulate species-specific pests favouring greater investment in defence traits. Yet it is unknown how altered plant rhizosphere interactions influence the plant diversity-driven selection for altered plant phenotypes.

2. We tested for plant diversity-driven selection on plant aboveground traits and how these traits are modified by their rhizosphere microbial communities after 11 years in experimental plant monocultures and mixtures. Plants propagated from monocultures or mixtures were grown in combination with their 'home' vs. 'away' arbuscular mycorrhizal fungi (AMF) or non-AMF microbes in two separate experiments using five and eight plant species respectively. We hypothesized plants in monocultures may be selected for better defence and better performance in association with rhizosphere microbial communities compared with plants in mixtures.

3. Monoculture and mixture plants significantly differed in their aboveground phenotypes. As predicted, plant traits related to defence (greater leaf mass per area and leaf dry matter content and reduced leaf damage) were more pronounced in monoculture plants in both experiments. Effects of the rhizosphere microbial communities, which generally enhanced plant growth, tended to be species-specific. We found interactive effects of diversity-driven selection for the AMF treatment in four plant species, indicating that plants selected in monoculture vs. mixture differed in their response to monoculture- vs. mixture-derived AMF communities.

47 **4.** We conclude that long-term differences in plant diversity lead to selection for altered plant
48 phenotypes. Such differences may be further modified in association with the AMF microbial
49 communities derived from the different plant diversity treatments, but often outcomes are
50 species-specific. This suggests that plant species differ in their capacity to respond to
51 diversity loss and associated changes in rhizosphere microbial communities, making it
52 complicated to predict community-level responses to such loss.
53
54 ***Keywords***: Arbuscular mycorrhizal fungi, Biodiversity experiment, Natural selection, Non-
55 AMF root-associated microbes, Phenotypic variation, Plant–soil feedbacks, Rapid evolution,
56 Reciprocal transplant design
57

1 | INTRODUCTION

Plants engage in many ecological interactions throughout their life cycle that influence their phenotype and fitness (Schmid, 1992; Ackerly et al., 2000; Dudenhöffer et al., 2018). These ecological interactions include competitive interspecific interactions among plant species, as well as antagonistic and facilitative interactions with the soil microbes they accumulate. Such interactions are the main ecological determinants of the composition and performance of plant species within a community (HilleRisLambers et al. 2012; van der Putten et al. 2013). Over time, selection pressure from interspecific interactions among plants and interactions with soil microbes may select for plants with traits favouring coexistence and altering responses to local soil biota (Helgason & Fitter, 2009; terHorst & Zee, 2016). For instance, species-rich communities can select for character displacement and increased division of labor among plant species (Zuppinger-Dingley et al., 2014; van Moorsel et al. 2018). Additionally, interactions with soil organisms can alter plant traits (Streitwolf-Engel et al., 1997; Goverde et al., 2000; Smith & Read, 2008) and influence plant fitness (Dudenhöffer et al., 2018). Specifically, interactions between plants and their root-associated microbes have the potential to incur selection on plants as well as the composition and functioning of soil microbial communities (Lau & Lennon, 2012; Schweitzer et al., 2014; Wagg et al., 2015; terHorst & Zee, 2016). However, it is unknown to which extent differences in selection between plant monocultures vs. mixtures may be influenced by co-occurring rhizosphere microbial communities, such as arbuscular mycorrhizal fungi (AMF) or soil bacteria.

Greater plant species richness is known to improve net biomass production in experimental communities and this effect was observed to strengthen over time (Cardinale et al., 2007; Allan et al., 2011; Reich et al., 2012; Meyer et al., 2016; Guerrero-Ramírez et al., 2017; Huang et al., 2018). A proposed mechanism behind this temporally strengthening

richness–productivity relationship is the decline in productivity in monocultures over time due to the accumulation of pests (Eisenhauer et al., 2012; Kulmatiski et al., 2012; Marquard et al., 2013; Schnitzer et al., 2011; van der Putten et al., 2013; Meyer et al. 2016). If pathogen and pest accumulation in monocultures are responsible for declining productivity over many years, selective pressure may favour the persistence and fitness of individuals with a greater tolerance and ability to benefit from their associated rhizosphere microbial communities and greater investment in defence traits, such as tougher and denser leaf tissues (Agrawal & Fishbein, 2006; Hanley et al., 2007; Zuppinger-Dingley et al., 2016).

Nevertheless, species-rich plant communities may maintain greater productivity by promoting greater soil biodiversity and thus the dilution of species-specific pathogens (Burrows & Pfleger, 2002; König et al., 2010; Scherber et al., 2010; Milcu et al., 2013). Additionally, species-rich plant communities may favour species that vary more in traits that capture resources in space and time to avoid interspecific competition and therefore more fully use the local resource pool (Roscher et al., 2013; Meyer et al., 2016; Cadotte, 2017; Wagg et al., 2017). Interspecific competition at high diversity may not only favour species with complementary functional traits but may also select for increased complementarity between species via evolutionary changes within species. For example, character displacement leading to increased complementarity and facilitation has been observed in a long-term field biodiversity experiment (Zuppinger-Dingley et al., 2014; Schöb et al., 2018).

Here we address whether selection in plant communities driven by low vs. high plant species richness (i.e. monoculture vs. mixture) results in altered traits and how these phenotypic responses to selection may be modified by the different rhizosphere microbiomes which developed in association with the respective plant communities. This effect of species diversity loss resulting in different phenotypes we refer to as “diversity-driven selection”. For instance, differences in plant phenotype between monocultures vs. mixtures may result from

differential survival, growth and reproduction of genotypes (see Fakheran et al., 2010), from recombination during sexual reproduction and from new mutations, presumably with decreasing order of importance in perennial plant communities observed over limited time spans (van Moorsel et al. 2018). Based on the arguments presented above, we hypothesize that (1) monoculture plant phenotypes may be selected for better defence —assessed here by greater leaf mass per area (LMA) and leaf dry matter content (LDMC); and lower leaf damage caused by pests compared with mixtures. We further hypothesized that (2) plants selected in monoculture would perform better (greater biomass and investment in defence traits with less aboveground pest damage) than plants selected in mixture when associated with the rhizosphere microbial communities from the corresponding species' monoculture. Finally, we hypothesized that (3) plants selected in mixture or monoculture would do better with their respective mixture- or monoculture-derived microbial communities ("home" combination) than with their non-matching rhizosphere microbial-community treatments ("away" combination).

2 | MATERIALS AND METHODS

2.1 | Conditioning of plants and soil communities

We tested our hypotheses using 11-year old plant monocultures and mixtures from the long-term Jena Experiment where plant community composition and diversity has been shown to determine the assemblage of AMF and rhizosphere microbial communities (Dassen et al., 2017, Schmid et al., 2019). The Jena Experiment was set up in 2002 to assess the effects of plant species richness on ecosystem functioning. It is located near the Saale River in Jena, Germany, and is described in detail elsewhere (Roscher et al., 2004; Weisser et al., 2017). In brief, plots were sown with differing plant species richness (1, 2, 4, 8, 16 or 60) and different compositions of plant functional groups (grasses, legumes, tall herbs and small

herbs). The sown plant community composition was maintained by manual weeding every spring, summer and fall since 2002. For our study multiple steps were taken to ensure that we had progeny plants produced from the plant populations that had experienced selection pressure from plant monocultures or (four- to) eight-species mixtures and passed through at least two sexual reproduction events (Fig. 1). The latter was done to allow for the possibility of recombination; otherwise, evolution in these perennial plants would have been largely restricted to "genotype sorting" (van Moorsel et al., 2018). In addition, using seed material for the present experiments was also done to reduce maternal environmental effects. From the initial sowing of plots in 2002 up to 2010 natural selection processes (Fig. 1, Step 1) led to the evolution of different phenotypes in plant monocultures and mixtures (Zuppinger-Dingley et al., 2014).

In spring 2010 (Fig. 1, Step 2) we collected plant cuttings from 48 plots of the Jena Experiment (12 monocultures, 12 two-species mixtures, 12 four-species mixtures and 12 eight-species mixtures). The cuttings were propagated in a glasshouse and then transplanted into an experimental garden in Zurich, Switzerland, in identical species composition as in the Jena Experiment, to obtain seeds from controlled pollination (for details see Zuppinger-Dingley et al., 2014). While plants were collected and propagated in Zürich, soils from the monoculture and mixture plots were exhumed to a depth of 30 cm, mixed and replaced into 1 x 1 m subplots within the original plots as previously described in Schmid et al. (2019). This mixing was done to provide soil containing the entire microbial communities that had developed in the rhizosphere of the different plant monocultures and mixtures in each 1 x 1 m subplot.

In spring 2011 (Fig. 1, Step 3), the seeds were germinated in a glasshouse in Zurich and then transplanted back in the same species composition into the 1 x 1 m subplots of the original plots in the Jena Experiment in Step 2. The new communities were maintained for

158 another three years until 2014 to allow the plants to further experience selection in
159 monocultures and mixtures and to re-assemble their own rhizosphere microbial communities
160 in the mixed soil (Schmid et al., 2019).

161 In March 2014 (Fig. 1, Step 4), we again collected plants from the 1 x 1 m subplots to
162 establish 48 communities of identical species composition as in Steps 1–3 in the common
163 garden in Zurich. These communities were only used for a second controlled sexual
164 reproduction event by fencing and allowing pollinators to freely fly within communities
165 (Schmid et al., 2019). The seeds from this second controlled sexual reproduction were then
166 used to establish the pot experiments shown in Step 6 in Fig. 1.

167 Before we replanted the 48 communities in Zurich (see Step 4), we removed ca. 100 g
168 of soil from the rhizosphere of several plant individuals from each monoculture plot and of
169 several plant individuals from four- or eight-species subplots in Jena (Table 1; Schmid et al.,
170 2019). By 2014, these soil communities had eight years to establish with respective plant
171 communities (Step 1), were then mixed, and could re-associate with their original plant
172 communities for three years. The rhizosphere soils were pooled across plant individuals and
173 then sieved separately for each species and subplot through a 5-mm mesh to homogenize the
174 soil. These soil samples were then used in trap-cultures to create the AMF and non-AMF
175 inoculum for the two experiments described below (Fig. 1, Step 5).

176 The plants used in the pot experiments (Fig. 1, Step 6) were derived from populations
177 that had undergone at least two generational turnover events (Step 2 and 4) and experienced
178 diversity-driven selection pressure in monoculture or mixture for a total of 11 years (Steps 1
179 and 3). Seeds were collected in summer 2014 from the plots established in Zurich. We
180 included only five and eight species for Experiment 1 and Experiment 2, respectively, as
181 plant individuals were needed from both monoculture and mixture selection history. The
182 species chosen are known to form mycorrhizal associations (Harley & Harley, 1987). To

183 avoid potential contamination with microbes attached to the seeds, all seeds were first surface
184 sterilized with 7–14% bleach and thoroughly rinsed before germination on 1% water-agar at
185 room temperature under natural light conditions.
186
187 **2.2 | Experiment 1: AMF-community effects on diversity-selected plants**
188 We processed 25 g of fresh rhizosphere soil removed from the rhizosphere of the
189 different plant species of each plant community through a series of sieves from 500 to 32 µm
190 using sugar gradient-centrifugation (Sieverding, 1991). From each sample we then collected
191 300–400 AMF spores under a dissecting microscope using 200-fold magnification. The
192 spores were placed in 30 ml of deionized water and then transferred to a 2-L trap-culture pot
193 containing autoclaved (120 °C for 99 min) 4:1 sand–soil mixture. Seeds, obtained from the
194 original supplier of the seeds used to establish the Jena Experiment (Rieger-Hofmann GmbH,
195 Blaufelden-Raboldshausen, Germany), were surface sterilized with 7–14% bleach and
196 germinated on 1% water-agar. Several plantlets of a single species were then planted into a
197 pot containing AMF spores derived from the Jena Experiment collected from the same plant
198 species. We included negative control trap-cultures of 30 ml of deionized water with no AMF
199 spores. Two trap-culture pots were cultivated per plant species with monoculture or mixture
200 history. Thus, there were two inoculum sources per plant species, one propagated from the
201 species monoculture and one propagated from a plant community that contained the species
202 within the mixture. Trap-cultures were cultivated over 10 months under glasshouse
203 conditions, after which the pots were allowed to dry out to encourage spore production. Prior
204 to ceasing watering, a root sample from each trap culture and control-trap culture was
205 collected and fixed in 50% ethanol, cleared with 10% KOH and stained with 5% ink-vinegar
206 (Vierheilig et al., 1998). The percentage of AMF root colonization was determined using the
207 transect-intersect method (McGonigle et al., 1990). We isolated AMF spores from trap-

208 cultures using the same method as described above. Five of the eight plant species had
209 sufficient AMF colonization and spores in trap-cultures from both plant monocultures and
210 mixture history: three small herbs (*Plantago lanceolata* L., *Prunella vulgaris* L. and *Veronica*
211 *chamaedrys* L.), one tall herb (*Galium mollugo* L.), one legume (*Lathyrus pratensis* L.). The
212 inoculum was prepared by cutting the dried roots into small fragments and homogenizing
213 them with the trap-culture substrate. We included a negative AMF control using the control
214 trap culture substrate (negative control) and a positive AMF control using *Rhizoglomus*
215 *irregulare,* a common AMF in grasslands known to heavily colonize plants and influence
216 growth (e.g. Wagg et al., 2011). *Rhizoglomus irregulare* was obtained from M.G.A. van der
217 Heijden (Agroscope Reckenholz-Tänikon, Zurich, Switzerland) and had no shared history
218 with plants or soils from the Jena Experiment as it was cultured for nine months in a substrate
219 of 15% soil, 65 % sand and 20 % oil binder with *Plantago lanceolata* plants.
220 To test for the effects of diversity-driven selection and AMF microbial-community
221 (short AMF history) treatments, we set up a complete factorial design for each species where
222 plants selected in monoculture vs. mixture were reciprocally combined with AMF from plant
223 monocultures or mixtures (20 treatment combinations: five plant species by two plant
224 histories by two AMF histories). These treatments were replicated 10 times for a total of 200
225 pots. The negative and positive AMF controls resulted in an additional 20 treatment
226 combinations (two AMF controls by five plant species by two plant histories) that were
227 replicated five times for an additional 100 pots, resulting in a total of 300 pots (Table S1).
228 However, we did not have enough seedlings of *G. mollugo* plants selected in mixture and
229 therefore the AMF monoculture and mixture treatments were replicated 9 and 8 times,
230 respectively, resulting in 297 pots (Table S1). Pots were filled with gamma radiated 1:1
231 sand–soil mixture (*w*/*w*) and inoculated with 9% inoculum (*v*/*v*) from the trap-cultures by
232 mixing the inoculum throughout the sterile substrate. The field soil used originated from a

natural grassland located at the University of Zürich. A 9% inoculum was used to ensure that inoculum abiotic effects were minimal while providing enough propagules of the AMF microbial communities to establish the desired biological treatments (Brinkmann et al. 2010). Pots were planted with a single seedling and randomly arranged in five experimental blocks in a glasshouse compartment with all treatment combinations represented equally in each block.

To standardize the non-AMF component of the soil microbial community within each pot, a microbial wash was created using 1.2L unsterilized field soil, collected from a natural pasture in Zurich, passed through a series of sieves and finally through Macherey-Nagel MN615 filter paper with 5L of deionized water. Each pot received 10 ml of the microbial-wash filtrate in addition to the AMF-treatment inoculum.

2.3 | Experiment 2: non-AMF microbial-community effects on diversity-selected plants

The design of Experiment 2 paralleled that of Experiment 1, however, here a non-AMF microbial-community treatment replaced the AMF treatment and all eight plant species were included. A detailed description of the set-up of Experiment 2 is provided in Schmid et al. (2019), who analysed the rhizosphere microbiomes on the plants inoculated with the different non-AMF microbial communities. The non-AMF microbial inoculum was generated using the same fresh soils that were used to generate the AMF inocula in Experiment 1. From this soil we made a microbial wash using 500 ml of deionized water and 25g of fresh rhizosphere soil filtered down to a mesh size of 25 µm. By filtering the soil slurry to 25 µm we excluded AMF propagules as has been done in previous studies to assess effects of the soil microbiomes in the absence of AMF (Koide & Li, 1989; Schnitzer et al., 2011; Wagg et al., 2014). Trap cultures were set up in the same manner as in Experiment 1 using two microbial-wash trap-culture pots per plant species and plant history in 2-L pots filled with an

autoclaved 4:1 sand-soil mixture and included two negative control trap-culture pots that received an autoclaved mix of all microbial-wash treatments. Each trap-culture pot received 250 mL of microbial wash into which one seedling per pot was transplanted. After 10 months of establishment in the glasshouse, roots of the trap-culture plants were cut into small fragments (< 5 cm) to generate homogenous inoculum and the absence of AMF colonization of roots was confirmed by microscopy.

The experimental phase also paralleled Experiment 1. We again used 1-L pots with gamma-radiated 1:1 sand-soil mixture (*w*/*w*) and inoculated the soil with 9% (*v*/*v*) inoculum from the trap-cultures. This gave us a factorial design of three non-AMF microbial-inoculation treatments (microbial inoculum from plant monocultures or mixtures and a negative control inoculum), two plant histories (plants selected in monoculture vs. mixture) and eight plant species. Each combination was replicated seven times resulting in 336 pots (Table S1). Pots were randomly arranged within replicate blocks in the glasshouse.

2.4 | Data collection

In both experiments, plants were grown in glasshouse conditions for five months. After three months we harvested aboveground biomass at 4 cm above the soil surface (first biomass harvest). Before the final destructive harvest after five months of growth, we assessed the level of leaf damage caused by fungus gnats *(Bradysia* spp.), two-spotted spider mite (*Tetranychus urticae* Koch), white fly (family Aleyrodidae) and infection by powdery mildew (family Erysiphaceae). To capture the overall observed leaf damage by these common pests, we estimated the degree of leaf damage into six ranked categories from 0 (no damage) to 5 (heavily damaged).

We focus on aboveground traits known to be related to plant–plant interspecific competition and defence against herbivores. The following leaf traits were measured in both

283 experiments during the final harvest for each plant: leaf absorbance, leaf mass per area
284 (LMA), leaf dry matter content (LDMC). The leaf absorbance (SPAD-502Plus Chlorophyll
285 Meter, KONICA MINOLTA, INC., Osaka, Japan) was measured on three representative
286 leaves of each plant. We also measured the area and fresh weight of three representative
287 leaves (LI-3100C Area Meter, LI-COR, Lincoln, USA) immediately after harvesting. Finally,
288 we recorded the maximum height of each plant at harvest. The aboveground biomass was
289 then harvested at soil-level (second biomass harvest). The biomass of plant shoots and the
290 harvested leaves were dried at 70 °C for 48 h and weighed to determine LMA and LDMC
291 and aboveground biomass. For both experiments, roots were harvested, washed free of
292 adhering rhizosphere soil and cut into small 1–5 cm fragments. A random subsample of roots
293 was then stored in 50 % ethanol for assessing AMF root length colonization as described
294 above for assessing the success of the trap cultures in Experiment 1. All measurements are
295 listed in Table 1.
296
297 **2.5 | Data analysis**
298 The data of the two experiments were analysed separately using R, version 3.0.2 (R
299 Core Team, 2013). Not all plants survived until the end of the experiments and mortality was
300 noted at each harvest. We therefore assessed the variation in plant survival for each
301 experiment at each harvest using logistic models and analysis of deviance with block, plant
302 history (plants selected in monoculture vs. mixture), soil history (Experiment 1: four AMF-
303 inoculation treatments; Experiment 2: three non-AMF microbial-inoculation treatments),
304 plant species identity and interactions of the latter three as explanatory terms. Although plant
305 biomass was collected at two different harvests, the two were highly correlated with each
306 other (Experiment 1: Spearman rho = 0.790, $P < 0.001$, Experiment 2: Spearman rho = 0.565,
307 $P < 0.001$) and analysing harvests separately produced similar results. For this reason the

biomass values of these two harvests were summed and analysed as the net aboveground biomass per pot during the five months of growth. Aboveground biomass, morphological trait measurements, leaf damage estimates and AMF colonization (Experiment 1 only) were analysed using general linear models (Schmid et al. 2017). The explanatory terms of the models were the same as above, i.e. block, plant history, soil history, plant species identity and interactions. The terms for the soil-history treatments were partitioned into contrasts. In Experiment 1 the first contrast term compared control with AMF inoculation, the second compared *R. irregulare* with AMF inoculation from the Jena Experiment and the third compared inoculation with AMF derived from plant monocultures vs. mixtures. In Experiment 2 the first contrast term compared control to the non-AMF microbial inoculation and the second compared the inoculation with non-AMF microbes derived from plant monocultures vs. mixtures. By first partitioning out the variation explained by the control treatment, the remaining variation between the two inoculum histories could also be analysed in interaction with plant history. This interaction corresponds to a comparison between 'home' and 'away' combinations typical for reciprocal transplant experiments (Joshi et al., 2001). In Experiment 1, biomass and height were square-root transformed and in Experiment 2 the LDMC was log-transformed to meet ANOVA assumptions. Because not all plants were planted on the same day in Experiment 2, the number of days between planting and the final harvest was included as a covariate in the analysis. For Experiment 1, we assessed the relationship between aboveground biomass and AMF colonization using linear regression.

3 | Results

3.1 | Experiment 1: Effects of plant and AMF-community history

Plant survival — Of the 297 plants initially planted, 227 survived at the end of Experiment 1 (76%). Plants selected in monoculture vs. mixture differed in survival depending on plant

333 species (Tables S1, S2). *Galium mollugo* monoculture plants showed increased survival (93
334 %) compared with mixture plants (63%), but conversely *V. chamaedrys* monoculture plants
335 showed decreased survival (60 %) compared with mixture plants (83 %). There were no
336 differences in plant survival between AMF treatments (see Table S2).
337 *AMF colonization* — No AMF colonization was observed with the negative control
338 inoculum. Inoculation with AMF resulted in root colonization levels that varied among plant
339 species and inoculum treatments (Table S3). Overall, *R. irregulare* gave the highest level of
340 root colonization. The AMF from plant monocultures and mixtures produced similar
341 colonization levels in *L. pratensis* and *V. chamaedrys*, but *G. mollugo* produced lower levels
342 of root colonization with AMF from its monoculture than from the plant mixture (Fig. S1).
343 *Aboveground biomass and plant traits* — The effects of plant history on aboveground
344 biomass differed between plant species (Table 2, Fig. 2a). In *G. mollugo* monoculture plants
345 produced greater aboveground biomass than mixture plants, whereas *P. lanceolata* and *V.*
346 *chamaedrys* showed the opposite response. Effects of soil history, i.e. AMF treatments, on
347 aboveground biomass also differed between plant species (Table 2). This was driven by *G.*
348 *mollugo* and *P. vulgaris* producing low biomass in the non-AMF control and high biomass
349 with *R. irregulare* (Fig. 2d, Table 2). However, aboveground biomass was similar for plants
350 inoculated with AMF derived from plant monocultures and mixtures in all species.
351 As with biomass, LDMC also showed plant species-specific responses to the plant
352 history and AMF treatments (Table 2). *Lathyrus pratensis* and *P. lanceolata* monoculture
353 plants had lower LDMC than mixture plants whereas the opposite was found in *V.*
354 *chamaedrys* (Fig. 2b). Furthermore, plants growing without AMF had particularly high
355 LDMC values in *L. pratensis* (Fig. 2e). Averaged across all species leaf damage was lower
356 for monoculture plants than for mixture plants, and this was particularly strong in *P.*
357 *lanceolata* (Fig. 2c). Differences in leaf damage among AMF treatments and corresponding

358 interaction terms were not significant, although *G. mollugo* had highest levels of leaf damage
359 in the absence of AMF (no-AMF control), which was opposite to the response in *L. pratensis*
360 (Fig. 2f).
361 Plant height, LMA and leaf absorbance were significantly affected by the three-way
362 interaction of plant history, AMF treatment and plant species identity (Table 2). Overall,
363 averaged across all plant species and AMF treatments plants selected in monoculture tended
364 to grow taller with lower LMA and showed lower leaf absorbance than plants selected in
365 mixture. However, the interaction between plant history and the two AMF inoculations
366 derived from plant monocultures vs. mixtures and plant species identity indicated that home-
367 vs.-away effects occurred in some species (Fig. 3). Thus, some species altered the expression
368 of these traits depending on whether their history matched that of the history of the AMF
369 inoculum. In *V. chamaedrys,* home combinations had taller plants with lower LMA than
370 away combinations (Fig. 3c). *Lathyrus pratensis* home combinations showed lower leaf
371 absorbance than away combinations. The opposite pattern was observed for *P. vulgaris* (Fig.
372 3c).
373 *Relationship between aboveground biomass or plant traits and AMF colonization* — We
374 found that greater AMF root colonization was correlated with greater aboveground biomass
375 in *G. mollugo*, *L. pratensis* and *P. vulgaris* but not in *P. lanceolata* and *V. chamaedrys* (Fig.
376 S3). Beyond this, plant traits were not correlated with AMF colonization, except for a
377 positive correlation between leaf absorbance and AMF colonization in *L. pratensis*. All
378 relationships between aboveground biomass or plant traits and AMF colonization were
379 independent of plant history and AMF treatment.
380

3.2 | Experiment 2: Effects of plant and non-AMF microbial-community history

Plant survival — Out of 336 plants initially planted, 232 survived at the end of Experiment 2 (69%, see Table S1). Plants from mixtures had higher survival than monoculture plants for all species in Experiment 2. Furthermore, survival differed significantly among non-AMF microbial treatments depending on plant species (Table S2). This interaction was driven by the control inoculum reducing the survival of *P. lanceolata*, where eight of 14 plants receiving the sterile inoculum did not survive.

Aboveground biomass and plant traits — Plant history had no overall and no interactive effects on aboveground biomass in Experiment 2 (Table 3, Fig. 4a). However, biomass was lower in the sterile control than in the other non-AMF microbial inoculated treatments in *G. mollugo*, *G. pratense*, *P. lanceolata* and *P. vulgaris* (Fig. 4g).

Independent of soil history, plant height, LMA and leaf absorbance were significantly affected by plant history in interaction with plant species identity (Table 3). Plants selected in monoculture were taller than plants selected in mixture in *Festuca rubra* and *V. chamaedrys*, while the opposite was the case for *G. mollugo* and *L. pratensis* (Fig. 4b). In contrast to the results of Experiment 1 but in agreement with our hypothesis, in Experiment 2 LMA was generally larger for plants selected in monoculture than in mixture and this effect was particularly strong in *F. rubra* (not included in Experiment 1) and *V. chamaedrys* (Fig. 4c). LDMC was differently affected by plant history and soil history depending on plant species identity (Fig 4i). For *L. pratensis* (different from Experiment 1) and *V. chamaedrys* (same as in Experiment 1) plants selected in monoculture had higher LDMC than plants selected in mixture (Fig. 4d). Furthermore, the two legumes *L. pratensis* and *O. viciifolia* had higher LDMC values when inoculated with non-AMF microbial communities compared to the control inoculum, and in particular those plants that had been selected in mixture (Fig. 4j). A three-way interaction between plant history, non-AMF microbial inoculation and plant

406 species occurred for LDMC, indicating that differences between monoculture and mixture
407 plants were particularly large when inoculated with the control microbial inoculum, but
408 varied in directions between the different plant species (Fig. S4). For *G. mollugo* and to a
409 small extent for *V. chamaedrys*, plants selected in monoculture had lower leaf absorbance
410 than plants selected in mixture (following the general trend of Experiment 1), but for all other
411 species plants selected in monoculture had higher values than plants selected in mixture (Fig.
412 4e).

413 As in Experiment 1, leaf damage was generally lower in plants selected in
414 monoculture than in mixture in Experiment 2, and this effect was again particularly strong in
415 *P. lanceolata* and additionally in *F. rubra* (Fig. 4f). Differences in leaf damage between non-
416 AMF microbial inoculation and corresponding interaction terms were not significant,
417 although the response of *F. rubra* and *G. mollugo* to the non-AMF microbial vs. control
418 inoculation treatments was opposite to the response of *G. pratense* and *P. vulgaris* (Fig. 4l).
419 Overall, we could not detect any differential effects between the non-AMF inocula generated
420 form plant monocultures vs. plant mixtures on aboveground biomass or plant traits (rows
421 with *MM* in Table 3, also see Fig. S5).

422

423 **4 | Discussion**

424 Using two glasshouse experiments with a parallel design we assessed whether (1)
425 plants selected in monoculture vs. mixture exhibited greater investment in leaf defence traits
426 and lower pest damage and (2) exhibit greater performance than plants selected in mixtures
427 when associated with the corresponding species' own monoculture rhizosphere microbial
428 communities. Furthermore, we tested (3) whether plants selected in monoculture would do
429 better with monoculture-derived microbial communities and plants selected in mixture would
430 do better with mixture-derived microbial communities than non-matching combinations.

Results from both experiments showed distinct phenotypic differences between plants selected in monoculture vs. mixture, but the strength and direction of the differences were often plant species-specific. Generally, however, the direction was in favour of greater leaf defence traits and reduced leaf damage in plants from monocultures in support of our hypothesis (1) suggesting greater selection for plant defence traits in monocultures compared with mixtures. While it has been hypothesized that selection of epigenetic variants may also be involved (Tilman & Snell-Rood, 2014), this was previously tested for in these plant communities and epigenetic changes were found to parallel genetic changes (van Moorsel et al., 2019). Although differential responses between plants selected in monocultures vs. mixtures to the rhizosphere microbial communities were observed, a greater performance of plants selected in monocultures vs. mixtures when associated with the corresponding species' own monoculture rhizosphere microbial community was rare, providing little support for our hypothesis (2). However, these plant history by rhizosphere community interactions did result from some home-vs.-away effects in combinations of plants selected in monoculture vs. mixture with AMF derived from monoculture vs. mixture providing some support for our hypothesis (3) that the matching of home-vs.-away plant and soil histories could provide the selected plants an advantage.

4.1 | Selection for increased defence in plant monocultures

In both Experiment 1 and 2 we found consistent results that *V. chamaedrys* plants selected in monoculture had greater LMA and LDMC than plants selected in mixture. However, in Experiment 1, the greater LMA of *V. chamaedrys* monoculture plants was only apparent when inoculated with an AMF community from plant mixtures. In Experiment 2 monoculture plants of *F. rubra*, *L. pratensis* and *P. vulgaris* showed greater LMA and LDMC

than mixture plants. These results support (1) that monocultures select for greater leaf defence such as leaf toughness, which is related to LDMC and LMA (Hanley et al., 2007; Pérez-Harguindeguy et al., 2013). Leaf toughness reduces digestibility, thereby protecting plants from herbivory (Turner, 1994). Increased leaf toughness has also been suggested to positively correlate with an accumulation of defence compounds in leaves (Coley, 1988). In our study higher LDMC and LMA in monoculture plants may suggest that in monocultures, plants are selected for increased leaf defence. Increased LDMC and LMA are additionally related to higher resource investment in leaves and longer foliar life span (Turner, 1994; Wilson et al., 1999; Westoby et al., 2002). Thus, the increased LDMC and LMA of monoculture plants in our study provide evidence that they have been selected for greater allocation of resources to leaves than their conspecifics in mixtures.

Leaf damage was also generally higher in plants with a mixture history than those from monocultures across all test species in both Experiment 1 and Experiment 2, most strongly in *P. lanceolata*. This further supports the concept that plants in monocultures likely experience selection favoring greater defence. *Plantago lanceolata* has often been used as a model species in grassland ecosystems to study plant responses to the local environment and ecological interactions in detail (Joshi et al., 2001). It has been shown that soil legacy effects can influence the expression of defence genes related to chewing herbivore defence in *P. lanceolata* (Zhu et al. 2018). Neighboring species and diversity have also been shown to influence defence-related leaf chemicals in the metabolome of *P. lanceolata* (Barton & Bowers 2006, Mraja et al. 2011).

4.2 | AMF-community effects on diversity-selected plants

The effect of different AMF treatments on plant phenotypes was species-specific,

479 confirming the plant species-specific context dependency of plant–AMF interactions (Bur-
480 rows & Pfleger, 2002; Hoeksema et al., 2010; Hoeksema et al., 2018). In addition, the degree
481 to which plants were affected by the associated AMF community not only varied among
482 plant species, but also depended on plant history, i.e. whether the inoculum of the AMF com-
483 munity was derived from the same plot as the plants ('home' versus 'away'). Such home-vs.-
484 away effects were observed for plant height, LMA and leaf absorbance. These home-vs.-
485 away effects were, however, inconsistent as sometimes away and sometimes home combina-
486 tions had higher trait values. For example, *G. mollugo* monoculture plants grew taller with
487 the AMF community from plant mixtures than with the AMF from the *G. mollugo* monocul-
488 ture, whereas *V. chamaedrys* in home combinations produced taller plants than in away com-
489 binations. Beneficial effects of AMF derived from plant mixtures may be due to the positive
490 effect of plant diversity on AMF community diversity (König et al., 2010), which in turn pro-
491 motes plant growth (van der Heijden et al., 1998; Wagg et al., 2011). Beneficial effects of
492 home combinations (see above examples of *V. chamaedrys* and *P. vulgaris*) suggest that for
493 these species a co-selection between plants and their AMF community may result in a change
494 of plant growth strategy. The development of positive plant–soil feedback effects over the
495 first 8 years of the Jena Experiment has previously been demonstrated (Zuppinger-Dingley et
496 al. 2016) and may have been due to such effects.
497 The different strength and directionality of the home-vs.-away effects of plant–AMF
498 combinations suggests that in the balance, co-selection between the two partners may in-
499 crease (home advantage) or decrease (home disadvantage) mutualistic interactions depending
500 on the species involved. Few studies have explicitly looked for effects of different types of
501 AMF inoculation on phenotypic traits of plants (Klironomos, 2003; Koch, Antunes, Maherali,
502 Hart, & Klironomos, 2017; Streitwolf-Engel, Boller, Wiemken, & Sanders, 1997), and even
503 fewer have done so for plants with different evolutionary history (Hoekssema et al. 2018).

For instance, Seifert et al. (2009) demonstrated that invasive North American populations of *Hypericum perforatum* had a finer root architecture and a reduced response to AMF inoculation than native European populations, thus revealing that the North American population had evolved an altered resource acquisition strategy. Next steps are to assess the AMF communities within the plant roots to further elucidate the mechanisms by which plants did or did not benefit from their own 'home' AMF partners.

4.3 | Non-AMF microbial-community effects on diversity-selected plants

Non-AMF rhizosphere microbial communities and their influence on plant growth can change over generations due to local natural selection processes (Lau & Lennon 2011; Wagg et al. 2015; Guamola et al. 2018). Therefore, inoculating plants with non-AMF microbial communities collected from plant monocultures should be more detrimental to plant growth if monocultures accumulate a greater abundance of pathogens. However, we did not detect main or interactive effects of the soil-history treatments using non-AMF microbial communities from plant monocultures versus mixtures here (see Table 3). In contrast to the expected overall negative effects, we found that in the absence of AMF, non-AMF microbes generally had an overall beneficial rather than detrimental effects on plant performance, as seen in the reduced aboveground biomass in the control treatment in Experiment 2.

The lack of non-AMF microbial-community effects on plant phenotypic traits was surprising because previously we found distinct differences in the rhizosphere microbiomes of our potted plants at harvest reflecting the different inocula from plant monocultures vs. mixtures in all plant species (Schmid et al. 2019). The only interactive effect on plant phenotypes we found was that the non-AMF microbial communities from the field increased LDMC in the two legume species *L. pratensis* and *O. viciifolia*. Legumes depend on rhizobia and may thus respond differently than other herbaceous species to these microbial

529 communities. It is known that legumes and their rhizobia may depend on genotype and
530 location (Denison & Kiers 2004; Kiers et al. 2010).
531
532 **5 | Conclusions**
533 Plant diversity has a positive long-term effect on a number of ecosystem properties
534 and in particular on the maintenance of plant productivity over time (e.g. Reich et al., 2012).
535 This may be due to on-going co-selection or assembly processes between different plant
536 species and between plants and their rhizosphere microbial communities. In species-rich
537 plant communities, greater productivity is likely due to increased complementarity among
538 species caused by rapid evolution of character displacement (Zuppinger-Dingley et al., 2014),
539 while increasing pest pressure may lead to a decline in monoculture productivity (Meyer et
540 al., 2016). Yet if both interspecific competition in mixtures and pest/pathogen resistance in
541 monocultures are strong selective forces, species should exhibit diversity-dependent
542 selection. Here we found evidence that plants from monocultures exhibit greater expression
543 of traits related to defence (higher LMA and LDMC and reduced leaf damage). Further we
544 found that for some of the tested plant species, the origin of AMF from plant monocultures
545 versus mixtures differentially affect plants selected in monoculture vs. mixture. This
546 emphasizes the need to further consider how species vary in their responses to selection
547 pressures induced by species loss. Only with such studies we can understand how diversity
548 loss may alter evolutionary trajectories of ecological interactions in plant communities.
549
550 **ACKNOWLEDGEMENTS**
551 We thank M. Furler, D. Topalovic, D. Trujillo Villegas and T. Zwimpfer for technical
552 assistance, D. Flynn, V. Yadav and D. Zuppinger-Dingley for the establishment of the field
553 plots and A. Ferrari, S. Karbin, J. Moser and Y. Xu for help with measurements. We are

554 grateful for the constructive comments of two anonymous reviewers on an earlier version of
555 the paper. This study was supported by the Swiss National Science Foundation (grants
556 number 147092 and 166457 to B. S.) and the University Research Priority Program Global
557 Change and Biodiversity of the University of Zurich. The Jena Experiment is supported by
558 the German Science Foundation (FOR 1451, SCHM 1628/5-2).
559

560 **AUTHOR CONTRIBUTIONS**

561 T.H., B.S., D.Z.D. and C.W. designed the experiment. T.H. and S.J.vM. performed the
562 experiment and collected the data. T.H., S.J.vM., M.W.S., B.S. and C.W. analysed the data
563 and produced the results. All authors contributed to the final written version of the
564 manuscript.
565

566 **DATA ACCESSIBILITY**

567 Data will be made publicly available on the datadryad.org upon acceptance of the manuscript.
568

569 **REFERENCES**

570 Ackerly, D.D., Dudley, S.A., Sultan, S.E., Schmitt, J., Coleman, J.S. Linder, R., … Lechow-
571 icz, M.J. (2000) The evolution of plant ecophysiological traits: recent advances and fu-
572 ture directions: new research addresses natural selection genetic constraints, and the
573 adaptive evolution of plant ecophysiological traits. *BioScience, 11*, 979-995.
574 Agrawal, A. & Fishbein, M. (2006) Plant defense syndromes. *Ecology, 87*, S132-S149.
575 Allan, E., Weisser, W., Weigelt, A., Roscher, C., Fischer, M. & Hillebrand, H. (2011) More
576 diverse plant communities have higher functioning over time due to turnover in comple-
577 mentary dominant species. *Proceedings of the National Academy of Sciences, USA, 108*,
578 17034-17039.

579 Barton, K.E. & Bowers, M.D. (2006) Neighbor species differentially alter resistance pheno-
580 types in Plantago. *Oecologia, 150*, 442-452.
581 Brinkman, P. E., van der Putten, W. H., Bakker, E. J., & Verhoeven, K. J. F. 2010. Plant-soil
582 feedback: Experimental approaches, statistical analyses and ecological interpretations.
583 *Journal of Eco*logy, 98, 1063–1073.
584 Burrows, R. L., & Pfleger, F. L. (2002). Host responses to AMF from plots differing in plant
585 diversity. *Plant and Soil*, *240*, 169–180.
586 Cadotte, M. W. (2017). Functional traits explain ecosystem function through opposing mech-
587 anisms. *Ecology Letters*, *20*, 989–996.
588 Cardinale, B.J., Wright, J.P., Cadotte, M.C., Carroll, I.T., Hector, A., Srivastava, D.S., Lo-
589 reau, M. & Weiss, J.J. (2007) Impacts of plant diversity on biomass increase through
590 time because of species complementarity. *Proceedings of the National Academy of Sci-*
591 *ences, USA, 104*, 18123-18128.
592 Coley, P.D. (1988) Effects of plant growth rate and leaf lifetime on the amount and type of
593 anti-herbivore defense. *Oecologia, 74*, 531-536.
594 Dassen, S., Cortois, R., Martens, H., de Hollander, M., Kowalchuk, G. A., van der Putten, W.
595 H., & De Deyn, G. B. (2017). Differential responses of soil bacteria, fungi, archaea and
596 protists to plant species richness and plant functional group identity. *Molecular Ecology*,
597 *26*, 4085-4098.
598 Denison, R.F & Kiers, E.T. (2004) Lifestyle alternatives for rhizobia: mutualism, parasitism,
599 and forgoing symbiosis. *FMES Microbiology Letters, 15*, 187-193.
600 Dudenhöffer, J.-H., Ebeling, A., Klein, A.-M., & Wagg, C. (2018). Beyond biomass: Soil
601 feedbacks are transient over plant life stages and alter fitness. *Journal of Ecology*, *106*,
602 230–241.

603 Eisenhauer, N., Reich, P. B., & Scheu, S. (2012). Increasing plant diversity effects on
604 productivity with time due to delayed soil biota effects on plants. *Basic and Applied*
605 *Ecology*, *13*, 571–578.
606 Fakheran, S., Paul-Victor, C., Heichinger, C., Schmid, B., Grossniklaus, U. & Turnbull, L.A.
607 (2010) Adaptation and extinction in experimentally fragmented landscapes. *Proceedings*
608 *of the National Academy of Sciences, USA, 107*, 19120-19125.
609 Goverde, M., van der Heijden, M. G. A., Wiemken, A., Sanders, I. R., & Erhardt, A. (2000). Arbuscu-
610 lar Mycorrhizal Fungi Influence Life History Traits of a Lepidopteran Herbivore. *Oecologia*,
611 *125*, 362–369.
612 Gomola, C.E., McKay, J.K., Wallenstein, M.D., Wagg, C. & O'Brien, M.J. (2018). Within-species
613 trade-offs in plant-stimulated soil enzyme activity and growth, flowering, and seed size. *Ecology*
614 *& Evolution, 8*, 11717-11724.
615 Guerrero-Ramírez, N.R., Craven, D., Reich, P.B., Ewel, J.J., Isbell, F., Koricheva, J. ... Eisenhauer,
616 N. (2017). Diversity-dependent temporal divergence of ecosystem functioning in experi-
617 mental ecosystems. *Nature Ecology & Evolution, 1*, 1639.
618 Hanley, M.E., Lamont, B.B., Fairbanks, M.M., Rafferty, C.M. (2007). Plant structural traits
619 and their role in anti-herbivore defense. *Perspectives in Plant Ecology, Evolution and*
620 *Systematics, 8*, 157–178.
621 Harley, J. L., & Harley, E. L. (1987). A check-list of Mycorrhiza in the British flora. *New*
622 *Phytologist*, *105*, 1–102.
623 Helgason, T. & Fitter, A. (2009). Natural selection and the evolutionary ecology of the arbus-
624 cular mycorrhizal fungi (*Phylum Glomeromycota*). *Journal of Experimental Botany, 60*,
625 2465–2480.
626 HilleRisLambers, J., Adler, P.B., Harpole, W.S., Levine, J.M. & Mayfield, M.M. (2012). Re-
627 thinking community assembly through the lens of coexistence theory. *Annual Review of*
628 *Ecology, Evolution, and Systematics, 43*, 227-248.

629 Hoeksema, J. D., Chaudhary, V. B., Gehring, C. A., Johnson, N. C., Karst, J., Koide, R. T.,
630 … Umbanhowar, J. (2010). A meta-analysis of context-dependency in plant response to
631 inoculation with mycorrhizal fungi. *Ecology Letters*, *13*, 394–407.
632 Hoeksema, J.D., Bever, J.D., Chakraborty, S., Chaudhary, V.B., Gardes, M., Gehring, C.A.,
633 … Zee, P.C. (2018). Evolutionary history of plant hosts and fungal symbionts predicts
634 the strength of mycorrhizal mutualism. *Communications Biology, 1*, 116.
635 Huang, Y., Chen, Y., Castro-Izaguirre, N., Baruffol, M., Brezzi, M., Lang, A., … Schmid, B.
636 (2018) Impacts of species richness on productivity in a large-scale subtropical forest ex-
637 periment. *Science, 362*, 80-83.
638 Joshi, J., Schmid, B., Caldeira, M. C., Dimitrakopoulos, P. G., Good, J., Harris, R., … others.
639 (2001). Local adaptation enhances performance of common plant species. *Ecology Let-*
640 *ters*, *4*, 536–544.
641 Kiers, E.T., Palmer, T.M., Ives, A.R., Bruno, J.F. & Bronstein, J.L. (2010) Mutualisms in a
642 changing world: an evolutionary perspective. *Ecology Letters, 13*, 1459-1474.
643 Klironomos, J. N. (2003). Variation in plant response to native and exotic arbuscular mycor-
644 rhizal fungi. *Ecology*, *84*, 2292–2301.
645 Koch, A. M., Antunes, P. M., Maherali, H., Hart, M. M., & Klironomos, J. N. (2017). Evolu-
646 tionary asymmetry in the arbuscular mycorrhizal symbiosis: conservatism in fungal mor-
647 phology does not predict host plant growth. *New Phytologist*, *214*, 1330–1337.
648 Koide, R.T. & Li, M. (1989). Appropriate controls for vesicular–arbuscular mycorrhiza re-
649 search. *New Phytologist, 111*, 35-44.
650 König, S., Wubet, T., Dormann, C. F., Hempel, S., Renker, C., & Buscot, F. (2010). TaqMan
651 real-time PCR assays to assess arbuscular mycorrhizal responses to field manipulation of
652 grassland biodiversity: effects of soil characteristics, plant species richness, and func-
653 tional traits. *Applied and Environmental Microbiology*, *76*, 3765–3775.

Kulmatiski, A., Beard, K. H., & Heavilin, J. (2012). Plant-soil feedbacks provide an additional explanation for diversity-productivity relationships. *Proceedings of the Royal Society B: Biological Sciences*, *279*, 3020–3026.

Lau, J. A., & Lennon, J. T. (2011). Evolutionary ecology of plant-microbe interactions: soil microbial structure alters selection on plant traits. *New Phytologist*, *192*, 215–224.

Marquard, E., Schmid, B., Roscher, C., De Luca, E., Nadrowski, K., Weisser, W. W., & Weigelt, A. (2013). Changes in the Abundance of Grassland Species in Monocultures versus Mixtures and Their Relation to Biodiversity Effects. *PLoS ONE*, *8*, e75599.

McGonigle, T.P., Miller, M.H., Evans, D.G., Fairchild, G. L., & Swan, J. A. (1990). A new method which gives an objective measure of colonization of roots by vesicular—arbuscular mycorrhizal fungi. *New Phytologist*, *115*, 495–501.

Meyer, S., Ebeling, A., Eisenhauer, N., Hertzog, L., Hillebrand, H., Milcu, A., ... Weisser, W. (2016). Effects of biodiversity strengthen over time as ecosystem functioning declines at low and increases at high biodiversity. *Ecosphere, 7*, e01619.

Milcu, A., Allan, E., Roscher, C., Jenkins, T., Meyer, S. T., Flynn, D., … Eisenhauer, N. (2013). Functionally and phylogenetically diverse plant communities key to soil biota. *Ecology*, *94*, 1878–1885.

Mraja, A., Unsicker, S.B., Reichelt, M.., Gershenzon, J. & Roscher, C. (2011). Plant community diversity influence allocation to direct chemical defense in *Plantago lanceolata*. *PLoS ONE, 6*, e28055.

Pérez-Harguindeguy, N., Díaz, S., Garnier, E., Lavorel, S., Poorter, H., Jaureguiberry, P., … Cornelissen, J.H.C. (2013). New handbook for standardised measurement of plant functional traits worldwide. *Australian Journal of Botany, 61*, 167-234.

R Development Core Team. (2013). R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.

679 Reich, P.B., Tilman, D., Isbell, F., Mueller, K., Hobbie, S.E., Flynn, D.F. & Eisenhauer, N.
680 (2012). Impacts of biodiversity loss escalate through time as redundancy fades. *Science,*
681 *336*, 589-592.
682 Roscher, C., Fergus, A.J.F. Petermann, J.S. Buchmann, N., Schmid, B. & Schulzed, E-.D.
683 (2013). What happens to the sown species if a biodiversity experiment is not weeded?
684 *Basic and Applied Ecology, 14*, 187-198.
685 Roscher, C., Schumacher, J., Baade, J., Wilcke, W., Gleixner, G., Weisser, W. W., …
686 Schulze, E.-D. (2004). The role of biodiversity for element cycling and trophic interac-
687 tions: an experimental approach in a grassland community. *Basic and Applied Ecology*,
688 *5*, 107–121.
689 Scherber, C., Eisenhauer, N., Weisser, W. W., Schmid, B., Voigt, W., Fischer, M., …
690 Tscharntke, T. (2010). Bottom-up effects of plant diversity on multitrophic interactions
691 in a biodiversity experiment. *Nature*, *468*, 553–556.
692 Schmid, B. (1992). Phenotypic variation in plants. *Evolutionary Trends in Plants, 6*, 45-60.
693 Schmid, B., Baruffol, M., Wang, Z.H. & Niklaus, P.A. (2017). A guide to analyzing biodiver-
694 sity experiments. *Journal of Plant Ecology 10*, 91–110.
695 Schmid, M.W., Hahl, T., van Moorsel, S.J., Wagg, C., De Deyn, G.B. & Schmid, B. (2019).
696 Feedbacks of plant identity and diversity on the diversity and community composition of
697 rhizosphere microbiomes from a long-term biodiversity experiment. *Molecular ecology,*
698 *28*, 863-878.
699 Schnitzer, S. A., Klironomos, J. N., HilleRisLambers, J., Kinkel, L. L., Reich, P. B., Xiao, K.,
700 … Scheffer, M. (2011). Soil microbes drive the classic plant diversity–productivity pat-
701 tern. *Ecology*, *92*, 296–303.
702 Schöb, C., Brooker, R.W. & Zuppinger-Dingley, D. (2018) Evolution of facilitation requires
703 diverse communities. *Nature Ecology & Evolution, 2,* 1381-1385

704 Schweitzer, J.A., Juric, I., van de Voorde, T.F.J., Clay, K., van der Putten, W.H. & Bailey, K.
705 (2014). Are there evolutionary consequences of plant–soil feedbacks along soil gradi-
706 ents? *Functional Ecology, 28*, 55-64.
707 Seifert, E.K., Bever, J.D. & Maron, J.L. (2009). Evidence for the evolution of reduced my-
708 corrhizal dependence during plant invasion. *Ecology, 90*, 1055-1062.
709 Sieverding, E., da Silva, G. A., Berndt, R., & Oehl, F. (2015). Rhizoglomus, a new genus of
710 the Glomeraceae. *Mycotaxon*, *129*, 373–386.
711 Smith, S. E., & Read, D. J. (2008). Mycorrhizal symbiosis (Third Edition). Academic Press,
712 Amsterdam.
713 Streitwolf-Engel, R., Boller, T., Wiemken, A., & Sanders, I. R. (1997). Clonal growth traits
714 of two prunella species are determined by co-occurring arbuscular mycorrhizal fungi
715 from a calcareous grassland. *Journal of Ecology*, *85*, 181–191.
716 terHorst, C.P. & Zee, P.C. (2016). Eco-evolutionary dynamics in plant-soil feedbacks. *Func-*
717 *tional Ecology, 30*, 1062-1072.
718 Tilman, D. & Snell-Rood, E.C. (2014) Ecology: Diversity breeds complementarity. *Nature*
719 *515*, 44-45.
720 Turner, I.M. (1994). Sclerophylly: primarily protective? *Functional Ecology, 8*, 669–675.
721 van der Heijden, M. G., Klironomos, J. N., Ursic, M., Moutoglis, P., Streitwolf-Engel, R.,
722 Boller, T., … Sanders, I. R. (1998). Mycorrhizal fungal diversity determines plant biodi-
723 versity, ecosystem variability and productivity. *Nature*, *396*, 69–72.
724 van der Putten, W. H., Bardgett, R. D., Bever, J. D., Bezemer, T. M., Casper, B. B., Fukami,
725 T., … Wardle, D. A. (2013). Plant-soil feedbacks: the past, the present and future chal-
726 lenges. *Journal of Ecology*, *101*, 265–276.

van Moorsel, S.J., Schmid, M., Hahl, T., Zuppinger-Dingley, D. & Schmid, B. (2018). Selection in response to community diversity alters plant performance and functional traits. *Perspectives in Plant Ecology, Evolution and Systematics, 33*, 51-61.

Van Moorsel, S.J., Schmid, M.W., Wagemaker C.A.M., van Gurp, T., Schmid, B. & Vergeer, P. (2019) Evidence for rapid evolution in a grassland biodiversity experiment. *Molecular Ecology*, *28*, 4097-4117.

Vierheilig, H., Coughlan, A. P., Wyss, U., & Piché, Y. (1998). Ink and Vinegar, a Simple Staining Technique for Arbuscular-Mycorrhizal Fungi. *Applied and Environmental Microbiology*, *64*, 5004–5007.

Wagg, C., Jansa, J., Schmid, B., & van der Heijden, M. G. A. (2011). Belowground biodiversity effects of plant symbionts support aboveground productivity. *Ecology Letters*, *14*, 1001–1009.

Wagg, C., Bender, S.F., Widmer, F., & van der Heijden, M.G.A. (2014). Soil biodiversity and soil community composition determine ecosystem multifunctionality. *Proceedings of the National Academy of Sciences, USA, 111*, 5266-5270.

Wagg, C., Boller, B., Schneider, S., Widmer, F. & van der Heijden, M.G.A. (2015). Intraspecific and intergenerational differences in plant–soil feedbacks. *Oikos, 124*, 994-1004.

Wagg, C., Ebeling, A., Roscher, C., Ravenek, J., Bachmann, D., Eisenhauer, N., Mommer, L., Buchmann, N., Hillebrand, H., Schmid, B. & Weisser, W. W (2017). Functional trait dissimilarity drives both species complementarity and competitive disparity. *Functional Ecology, 31*, 2320-2329.

Weisser, W., Roscher, C., Meyer, S.T., Ebeling, A., Luo, G., Allan, E., ... Eisenhauer, N. (2017). Biodiversity effects on ecosystem functioning in a 15-year grassland experiment: Patterns, mechanisms, and open questions. *Basic and applied ecology, 23*, 1-73

751 Westboy, M., Falster, D.S., Moles, A.T., Vask, P.A. & Wright, I. (2002). Plant ecological
752 strategies: some leading dimensions of variation between species. *Annual Review of*
753 *Ecology and Systematics, 33*, 125-159.
754 Wilson , P.J., Tompson, K. & Hodgson, J.G. (1999) Specific leaf area and leaf dry matter
755 content as alternative predictors of plant strategies. *New Phytologist, 143*, 155-162.
756 Zhu, F., Heinen, R., van der Sluijs, M., Raaijmakers, C., Biere, A. & Bezemer, T.M. (2018).
757 Species-specific plant-soil feedbacks alter herbivore-induced gene expression and de-
758 fense chemistry in *Plantago lanceolata*. *Oecologia, 188*, 801-811.
759 Zuppinger-Dingley, D., Flynn, D. F. B., De Deyn, G. B., Petermann, J. S., & Schmid, B.
760 (2016). Plant selection and soil legacy enhance long-term biodiversity effects. *Ecology*,
761 *97*, 918–928.
762 Zuppinger-Dingley, D., Schmid, B., Petermann, J. S., Yadav, V., De Deyn, G. B., & Flynn,
763 D. F. B. (2014). Selection for niche differentiation in plant communities increases biodi-
764 versity effects. *Nature*, *515*, 108–111.
765

766 **TABLE 1** Plant growth and leaf traits measured at the given time points in Experiment 1
767 ("Plant age" in weeks) and after 19–23 weeks in Experiment 2.

Plant species	**Functional group**	**Abbreviation**	
Festuca rubra†	Grass	*Fr*	
Plantago lanceolata	Forb	*Pl*	
Prunella vulgaris	Forb	*Pv*	
Veronica chamaedrys	Forb	*Vc*	
Galium mollugo	Forb	*Gm*	
Geranium pratense†	Forb	*Gp*	
Lathyrus pratensis	Legume	*Lp*	
Onobrychis viciifolia†	Legume	*Ov*	
Plant characteristic	**Units**	**Abbreviation**	**Plant age**
Total aboveground biomass	g/pot	*none*	12+20
Survival	yes / no	*none*	20
Colonization by AMF	%	*none*	20
Leaf absorbance	SPAD§	*none*	19
Leaf mass per area	µg/cm^2	LMA	20
Leaf dry matter content	g dry / g fresh	LDMC	20
Plant height (adjusted for biomass)	cm	Height	19
Leaf damage	0 – 5 (none - heavy)	*none*	20

768 Notes: † indicates species only present in Experiment 2. § SPAD values are index values,
769 defined by the manufacturer, which indicate the relative amount of chlorophyll present in the
770 leaf.
771

TABLE 2 ANOVA results for Experiment 1 assessing the effects of plant history (PH), species identity (Species or Sp) and AMF soil-inoculation treatments (Soil) and their interactions on plant performance and traits. Because pots were completely randomized within blocks (Block), all effects were tested against the residual. The overall variation among AMF soil-inoculation treatments is split into the indented contrast terms in italics, partitioning out the effects of the sterile control versus AMF present (*Str vs AMF* or *Str*), the inoculation with *R. irregulare* versus inoculation with AMF communities from the field (*Ri vs Field* or *Ri*), and AMF communities from the field cultivated under plant monocultures versus plant mixtures (*Mono vs Mix* or *MM*). For abbreviations of dependent variables see Table 1. *DF*, degrees of freedom; *%SS*, contribution to total sum of squares, i.e. percent variance explained; *P*, probability of type-I error; significant effects ($P<0.05$) are highlighted in bold.

		Aboveground biomass		Height		LMA		LDMC		Leaf absorbance		Leaf damage	
Source of variation	*DF*§	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*
Block	4	**5.36**	**<0.001**	0.77	0.071	**4.89**	**<0.001**	**6.05**	**0.001**	**3.76**	**<0.001**	**5.64**	**<0.001**
PH	1	0.04	0.527	**2.49**	**<0.001**	**0.88**	**0.014**	0.48	0.216	**0.46**	**0.018**	**1.46**	**0.015**
Species (Sp)	4	**64.90**	**<0.001**	**75.87**	**<0.001**	**59.82**	**<0.001**	**20.70**	**<0.001**	**76.95**	**<0.001**	**35.78**	**<0.001**
Soil	3	**2.10**	**<0.001**	0.27	0.386	0.60	0.247	0.78	0.480	0.34	0.238	0.43	0.616
Str vs AMF (Str)	1	**1.45**	**<0.001**	0.14	0.214	0.08	0.469	0.10	0.564	0.28	0.065	0.03	0.711
Ri vs Field (Ri)	1	**0.64**	**0.016**	0.12	0.241	0.33	0.130	0.58	0.174	0.05	0.439	0.38	0.212
Mono vs Mix (MM)	1	0.01	0.784	0.01	0.732	0.19	0.250	0.09	0.594	0.02	0.639	0.02	0.764
PH x Sp	4	**3.19**	**<0.001**	**0.88**	**0.043**	**1.56**	**0.032**	**3.28**	**0.037**	0.36	0.353	**6.66**	**<0.001**
PH x Soil	3	0.22	0.559	0.30	0.339	0.44	0.386	0.93	0.397	0.01	0.982	0.06	0.970
PH x Str	1	0.01	0.792	0.16	0.184	0.05	0.567	0.78	0.117	0.01	0.803	0.01	0.810
PH x Ri	1	0.20	0.170	0.10	0.281	0.03	0.644	0.11	0.552	0.00	0.867	0.01	0.808
PH x MM	1	0.01	0.745	0.04	0.509	0.36	0.115	0.05	0.704	0.01	0.780	0.03	0.721
Sp x Soil	12	**3.05**	**0.007**	**2.06**	**0.030**	2.36	0.189	**7.53**	**0.026**	1.23	0.239	3.14	0.372
Sp x Str	4	**2.37**	**<0.001**	0.79	0.065	0.41	0.592	**4.03**	**0.014**	0.03	0.986	1.35	0.235
Sp x Ri	4	0.25	0.676	0.47	0.260	1.26	0.073	1.54	0.302	**0.97**	**0.020**	1.31	0.249
Sp x MM	4	0.43	0.408	0.80	0.061	0.69	0.315	1.96	0.185	0.23	0.582	0.48	0.735
PH x Sp x Soil	11	1.37	0.320	1.30	0.201	**3.31**	**0.023**	3.54	0.423	**2.06**	**0.011**	2.59	0.465
PH x Sp x Str	3	0.20	0.599	0.15	0.633	0.24	0.643	0.23	0.868	0.20	0.472	1.01	0.246

PH x Sp x Ri	4	0.79	0.125	0.20	0.695	0.04	0.992	1.53	0.303	0.27	0.494	0.87	0.465
PH x Sp x MM	4	0.38	0.474	**0.95**	**0.031**	**3.03**	**0.001**	1.79	0.228	**1.58**	**0.001**	0.72	0.557
Residuals	184	19.78		16.07		26.14		56.71		14.83		44.23	

780 Note: § the residual *DF*s were 181 for LMA and LDMC due to plant mortality.
781

TABLE 3 ANOVA results for Experiment 2 assessing the effects of plant history (PH), species identity (Species or Sp) and non-AMF microbial soil-inoculum treatments (Soil) and their interactions on plant performance and traits. Because pots were completely randomized within blocks (Block), all effects were tested against the residual. However, because not all plants could be measured and harvested at the same time, we used a covariate (Days) to account for the number of number of days between planting and final harvesting. The overall variation among non-AMF microbial soil-inoculum treatments is split into the indented contrast terms in italics, partitioning out the effects of the sterile control versus inoculation with non-AMF microbial communities from the field (*Str vs Mic* or *Str*), and non-AMF microbial communities from the field cultivated under plant monocultures versus plant mixtures (*Mono vs Mix* or *MM*). For abbreviations of dependent variables see Table 1. *DF*, degrees of freedom; *%SS*, contribution to total sum of squares, i.e. percent variance explained; *P*, probability of type-I error; significant effects (*P*<0.05) are highlighted in bold.

		Aboveground biomass		Height		LMA		LDMC		Leaf absorbance		Leaf damage	
	Df§	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*	*%SS*	*P*
Block	6	**6.44**	**<0.001**	**2.87**	**<0.001**	**15.53**	**<0.001**	**7.79**	**<0.001**	**2.97**	**0.031**	**7.73**	**<0.001**
Days	1	**8.17**	**<0.001**	0.01	0.652	0.14	0.363	0.72	0.043	0.02	0.747	**4.37**	**<0.001**
PH	1	0.18	0.168	0.21	0.080	**1.76**	**0.002**	0.12	0.408	0.62	0.088	**1.54**	**0.002**
Species (Sp)	7	**55.44**	**<0.001**	**75.35**	**<0.001**	**32.35**	**<0.001**	**41.81**	**<0.001**	**45.88**	**<0.001**	**45.14**	**<0.001**
Soil	2	**2.58**	**<0.001**	0.30	0.108	0.05	0.860	0.21	0.543	0.36	0.420	0.12	0.677
Str vs Mic (Str)	1	**2.56**	**<0.001**	0.17	0.113	<0.01	0.891	0.13	0.385	0.10	0.499	0.08	0.474
Mono vs Mix (MM)	1	0.03	0.606	0.13	0.163	0.05	0.595	0.08	0.496	0.27	0.258	0.04	0.605
PH x Sp	7	0.74	0.355	**2.40**	**<0.001**	**4.49**	**0.001**	**2.75**	**0.031**	**5.70**	**<0.001**	1.00	0.482
PH x Soil	2	0.07	0.683	0.20	0.238	<0.01	0.990	0.52	0.225	0.28	0.508	0.14	0.640
PH x Str	1	0.04	0.517	0.00	0.874	<0.01	0.891	0.31	0.187	0.15	0.403	0.05	0.553
PH x MM	1	0.03	0.560	0.19	0.092	<0.01	0.964	0.22	0.265	0.14	0.419	0.08	0.462
Sp x Soil	14	**2.93**	**0.008**	1.38	0.128	3.37	0.160	**5.02**	**0.015**	3.17	0.245	1.66	0.697
Sp x Str	7	**1.84**	**0.008**	0.63	0.238	2.14	0.096	**3.88**	**0.003**	1.49	0.313	0.78	0.649
Sp x MM	7	1.09	0.122	0.75	0.139	1.23	0.422	1.14	0.480	1.67	0.245	0.88	0.570
PH x Sp x Soil	14	0.82	0.843	1.15	0.259	3.02	0.244	**5.38**	**0.009**	1.27	0.909	0.89	0.969
PH x Sp x Str	7	0.36	0.802	0.68	0.188	1.63	0.229	**3.80**	**0.004**	1.21	0.452	0.61	0.779
PH x Sp x MM	7	0.47	0.665	0.47	0.432	1.38	0.338	1.58	0.253	0.06	0.999	0.28	0.968

Residuals	†240	22.63	16.13	39.29	35.68	39.73	37.42

Note: § the residual *DF*s were 181 for LMA and LDMC due to plant mortality and 190 for Leaf absorbance because this was not measured on *F. rubra* due to its narrow leaf morphology, thus *DF*s or terms including Sp were 6 or 12 (instead of 7 or 14) for Leaf absorbance.

Figures

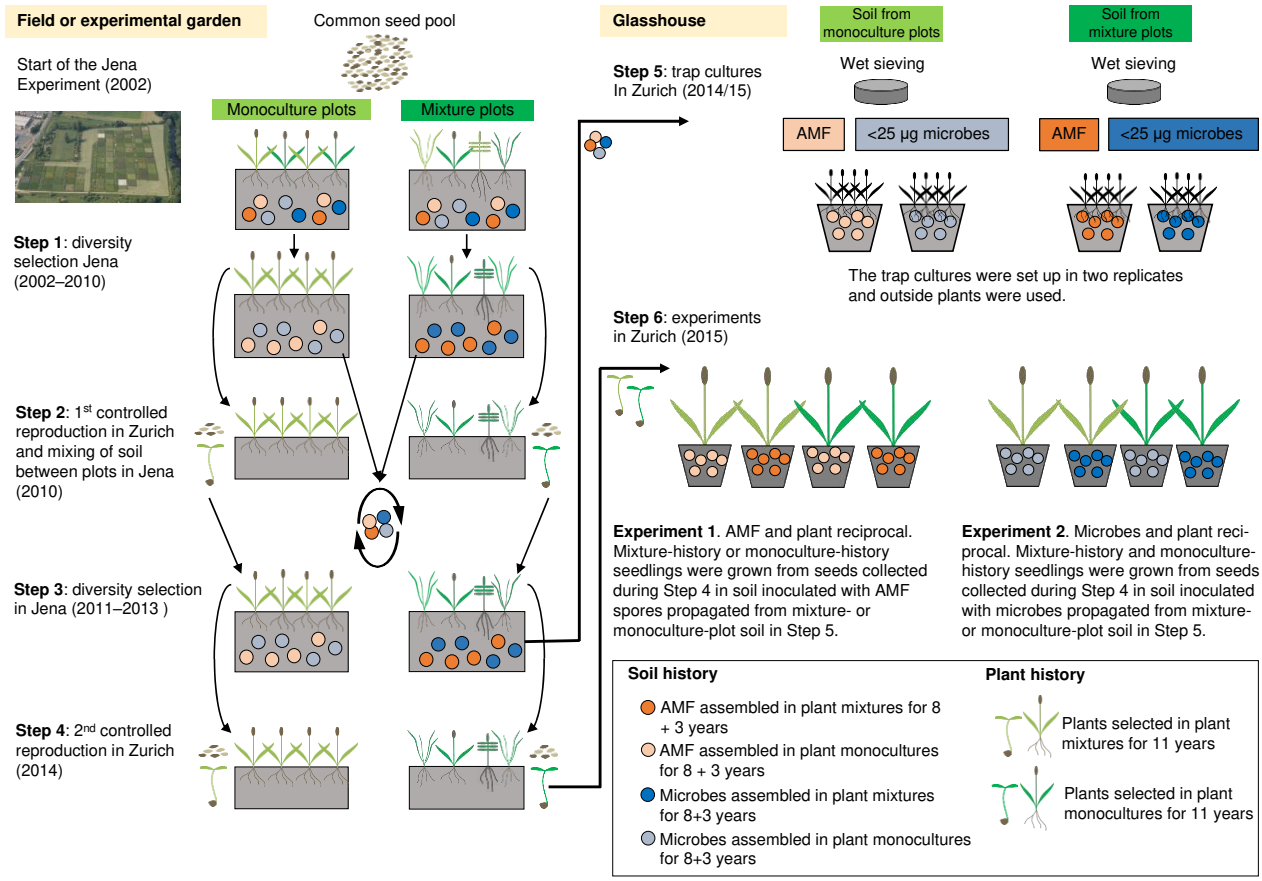


Figure 1. Schematic of the steps involved in the experimental design. The Jena Experiment was sown in 2002 using a common seed pool into monoculture and mixture plant communities. Plant material was collected in 2010 after 8 years of coexistence (Step 1). To ensure generational turnover within a species population and to produce propagules of each plant species population, plants were propagated in 2010 from cuttings and allowed to produce seeds within a common garden in Zürich (Step 2). These 2nd generation plants from the collected seeds were then transplanted back into the Jena Experimental field in their original compositions into a soil mixture from all plant monoculture and mixture plots to allow the progeny produced from the diversity selection from 2002–2010 to condition their own home-soil microbial communities (Step 3). Soils from the monoculture and mixture communities in Step 3 were collected in the fall of 2013 and used to produce AMF and non-

807 AMF soil microbial cultures within a standardized substrate and under controlled
808 environmental conditions in the glasshouse in Zürich (Step 5). This re-assembly of plant
809 communities using the 2nd generation plants (Step 2) also provided a second opportunity for
810 monocultures and mixture to impose selection on the 2nd generation of plants. The cuttings
811 from this 2nd generation were then again propagated in a common garden in Zürich to
812 produce seeds for a 3rd generation of the plant species populations for the current glasshouse
813 experiment (Step 6). See text and Schmid et al. (2019) for further explanation.
814

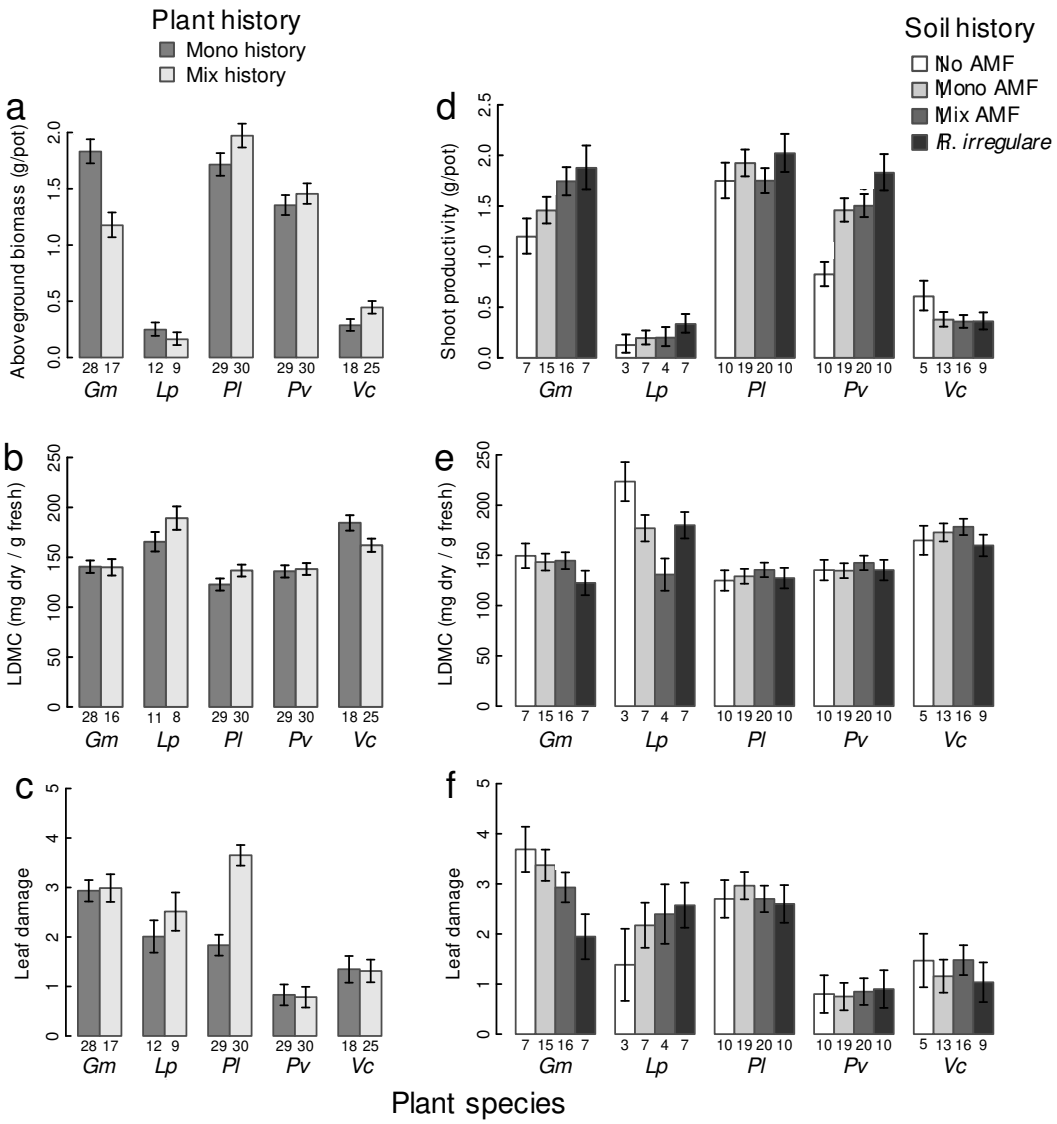
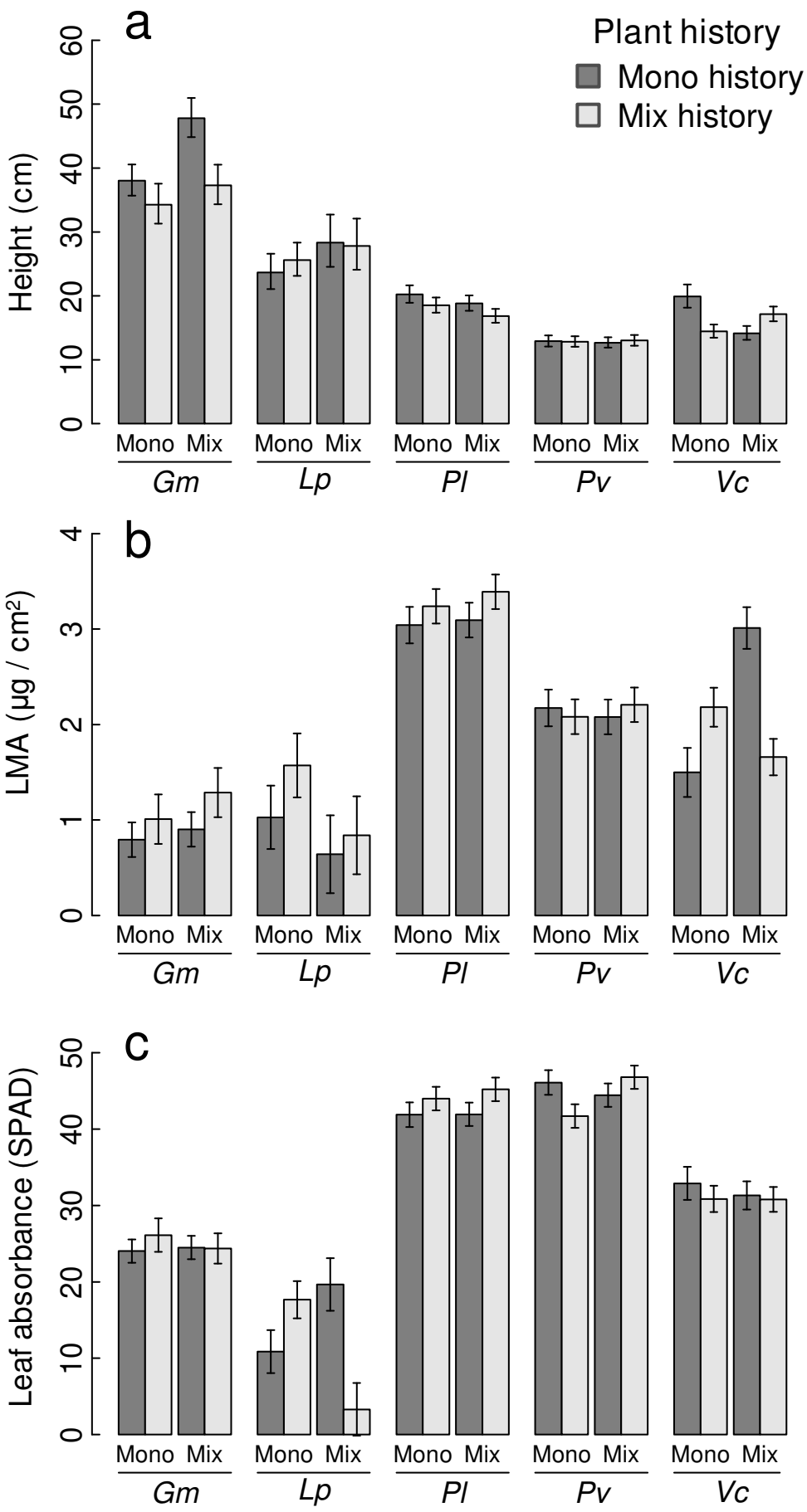
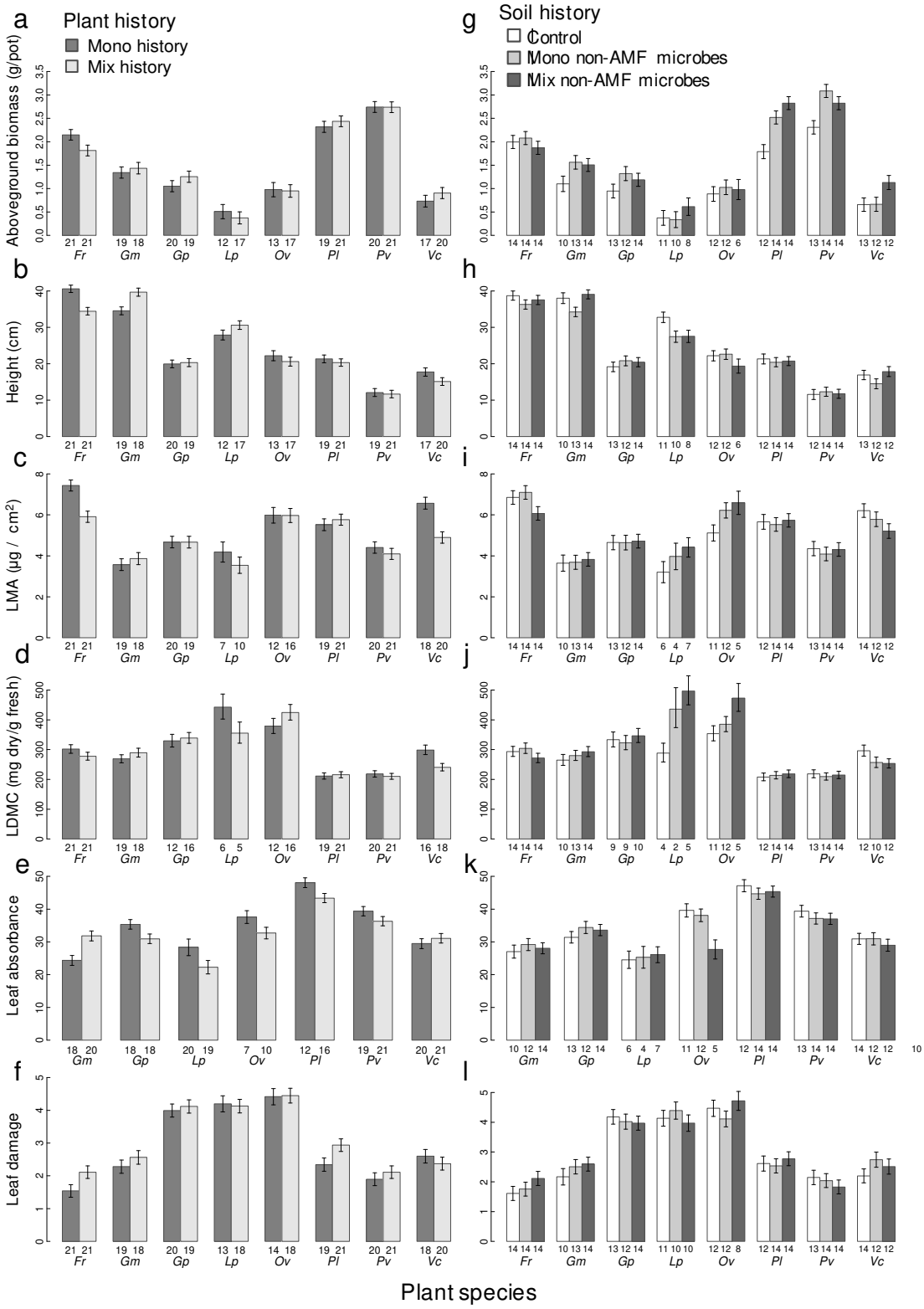


FIGURE 2 Total aboveground biomass (a, d), leaf dry matter content (LDMC; b, e) and leaf damage (c, f) of each plant species are shown for the two plant histories (plants selected in monoculture vs. mixture) in a–c) and the four soil histories (sterile inoculum, inoculation with *R. irregulare* and inoculation with AMF communities from the field cultivated under plant monocultures or plant mixtures in d–f) in Experiment 1. Error bars are standard errors of the model estimates and numbers below the means indicate the number of replicates for each. Plant species and trait abbreviations are defined in Table 1. Only aboveground biomass, LDMC and leaf damage are shown as these responses varied significantly among species by history and species by AMF treatment interactions (See Table 2 for significances).


a
Plant history
Mono history
Mix history
Height (cm)
0
10
20
30
40
50
60
Mono
Mix
Gm
Lp
Pl
Pv
Vc
b
LMA (µg / cm²)
0
1
2
3
4
c
Leaf absorbance (SPAD)
0
10
20
30
40
50

825

826 **FIGURE 3** Mean plant height, leaf mass per area (LMA), and leaf absorbance of each of the
827 five plant species illustrating significant three-way interactions of plant history (indicated by
828 shading of bars) x plant species identity (indicated below quartets of bars) x soil history (only
829 the two treatments with inocula from the Jena Experiment, indicated below pairs of bars) in
830 Experiment 1 (row *PH x Sp x MM* in Table 2). This three-way interaction can be understood
831 as species having different two-way interactions between plant and soil history, i.e. so-called
832 'home-vs.-away' effects (see text for further explanation). Error bars are standard errors of
833 the model estimates. Plant species and trait abbreviations are defined in Table 1. This three-
834 way interaction was not found to be significant for aboveground biomass, LDMC and leaf
835 damage (see Table 2), but is shown in Fig. S2.
836



837

838 **FIGURE 4** Total aboveground biomass (a, g), height (b, h), LMA (c, i), leaf dry matter con-

839 tent (LDMC; d, j), leaf absorbance (e, k) and leaf damage (f, l) of each plant species are

840 shown for the two plant histories (plants selected in monoculture vs. mixture) in a–f) and the

three soil histories (‘control’ microbial wash or soil microbes cultivated from plant monocultures or from plant mixtures) in g–l) in Experiment 2. See Table 3 for significance of species x plant history and species x soil history interactions. Error bars are standard errors of the model estimates and numbers below the means indicate the number of replicates for each. Plant species and trait abbreviations are defined in Table 1. Note that there was little evidence for three-way interactions (Table 3) and corresponding plots are thus only shown in Fig. S5.

Supplementary Materials

Table S1 Experimental design. Number of replicates for plants selected in monoculture vs. mixture (plant history) for each plant species grown in each soil-history treatment for both Experiment 1 (AMF inoculation) and Experiment 2 (non-AMF inoculation). Numbers in parentheses are the number of plants surviving to the end of the experiment. See main text for abbreviations and explanations of treatments.

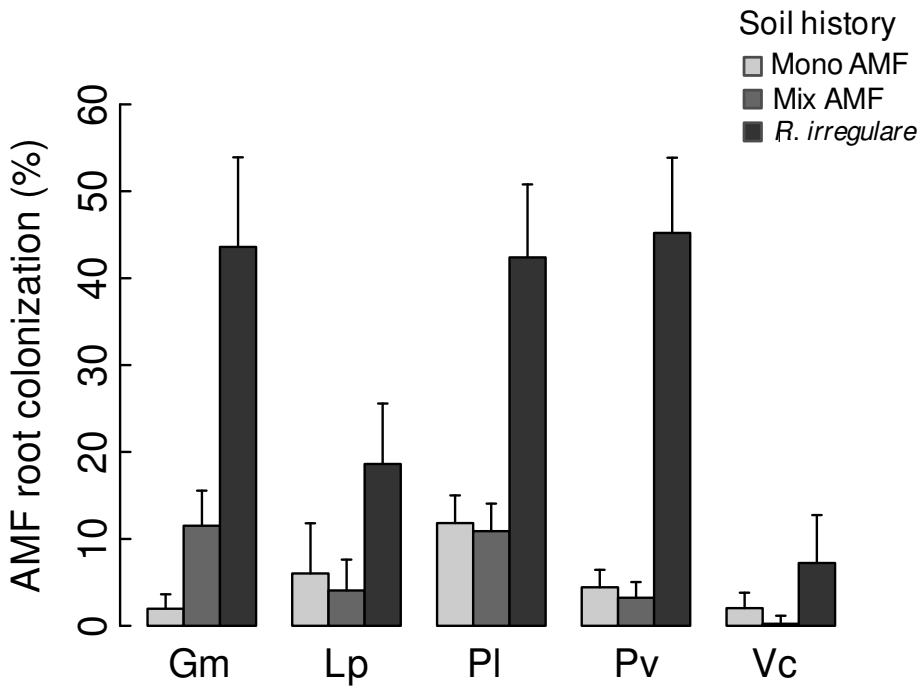
		Experiment 1				**Experiment 2**		
Species	Plant history	No AMF	Mono AMF	Mix AMF	*R. irreg.*	Sterile	Mono microbes	Mix microbes
Gm	Mono	5 (4)	10 (10)	10 (10)	5 (4)	7 (3)	7 (3)	7 (5)
Gm	Mix	5 (3)	9 (5)	8 (6)	5 (3)	7 (3)	7 (5)	7(4)
Lp	Mono	5 (3)	10 (3)	10 (2)	5 (4)	7 (5)	7 (3)	7 (4)
Lp	Mix	5 (0)	10 (4)	10 (2)	5 (3)	7 (6)	7 (6)	7 (4)
Pl	Mono	5 (5)	10 (9)	10 (10)	5 (5)	7 (3)	7 (7)	7 (6)
Pl	Mix	5 (5)	10 (10)	10 (10)	5 (5)	7 (3)	7 (6)	7 (7)
Pv	Mono	5 (5)	10 (9)	10 (10)	5 (5)	7 (6)	7 (7)	7 (7)
Pv	Mix	5 (5)	10 (10)	10 (10)	5 (5)	7 (7)	7 (7)	7 (7)
Vc	Mono	5 (2)	10 (5)	10 (7)	5 (4)	7 (5)	7 (4)	7 (5)
Vc	Mix	5 (3)	10 (8)	10 (9)	5 (5)	7 (7)	7 (7)	7 (6)
Fr	Mono	0	0	0	0	7 (7)	7 (7)	7 (7)
Fr	Mix	0	0	0	0	7 (7)	7 (7)	7 (7)
Gp	Mono	0	0	0	0	7 (4)	7 (2)	7 (3)
Gp	Mix	0	0	0	0	7 (1)	7 (4)	7 (4)
Ov	Mono	0	0	0	0	7 (2)	7 (2)	7 (1)
Ov	Mix	0	0	0	0	7 (4)	7 (3)	7 (2)

Table S2. Analysis of deviance assessing the effects of plant history (PH), soil history (Soil), species identity (Species or Sp) and their interactions on the survival of plants at the end of the experiments. Because pots were completely randomized within blocks (Block), all effects were tested against the residual. *DF*, degrees of freedom; *%Dev*, contribution to total deviance change, i.e. comparable to percent variance explained in ANOVAs (Tables 2 and 3 in main text); *P*, probability of type-I error based on quasi-F ratios of mean deviance changes; significant effects ($P<0.05$) are highlighted in bold.

	Experiment 1			Experiment 2		
Source of variation	*DF*	*% Dev*	*P*	*DF*	*% Dev*	*P*
Block	4	0.81	0.448	6	**3.66**	**0.005**
Plant history (PH)	1	0.03	0.689	1	**0.85**	**0.037**
Soil	3	1.44	0.088	2	0.31	0.444
Species (Sp)	4	**31.48**	**< 0.001**	7	24.83	**< 0.001**
PH x Soil	3	0.34	0.668	2	0.49	0.283
PH x Sp	**4**	**5.06**	**< 0.001**	7	1.65	0.288
Soil x Sp	12	3.85	0.131	14	**4.89**	**0.036**
PH x Soil x Sp	12	2.35	0.546	14	2.68	0.456
Residuals	252	54.65		282	60.63	

Table S3. ANOVA results for Experiment 1 assessing the effects of plant history (PH), species identity (Species or Sp) and soil history (AMF inoculation, Soil) and their interactions on the percent root length colonized by AMF. Because pots were completely randomized within blocks (Block), all effects were tested against the residual. The overall variation among soil-history treatments is split into the indented contrast terms in italics, partitioning out the effects of the sterile control versus AMF present (*Str vs AMF* or *Str*), the inoculation with *R. irregulare* versus inoculation with AMF communities from the field (*Ri vs Field* or *Ri*), and AMF communities from the field cultivated under plant monocultures versus plant mixtures (*Mono vs Mix* or *MM*). *DF*, degrees of freedom; *%SS*, contribution to total sum of squares, i.e. percent variance explained; *P*, probability of type-I error; significant effects ($P<0.05$) are highlighted in bold.

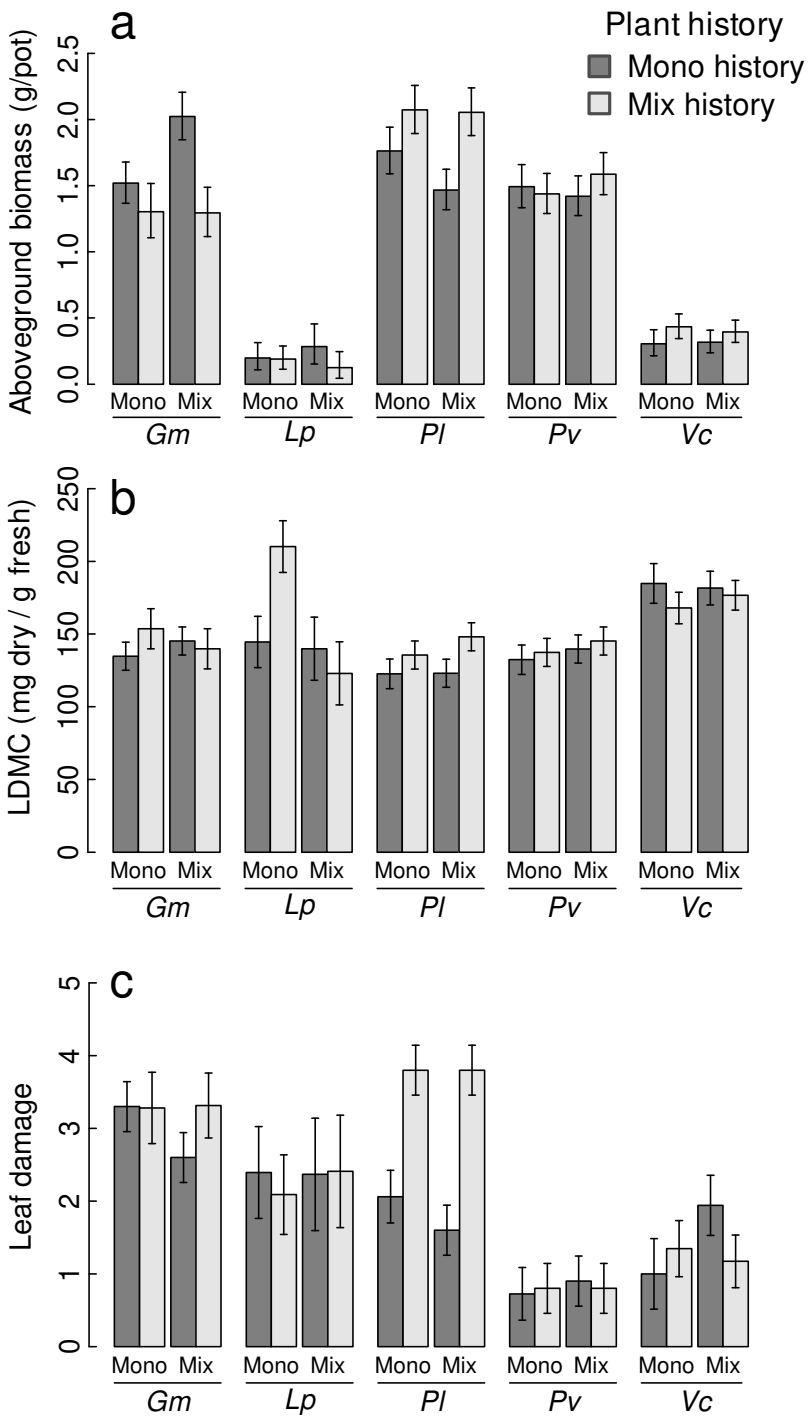
Source of variation	*DF*	*%SS*	*P*
Block	4	0.88	0.424
PH	1	0.01	0.825
Species (Sp)	**4**	**7.91**	**<0.001**
Soil	3	42.44	<0.001
Str vs AMF (Str)	**1**	**18.20**	**<0.001**
Ri vs Field (Ri)	**1**	**24.20**	**<0.001**
Mono vs Mix (MM)	1	0.04	0.671
PH x Sp	4	0.79	0.482
PH x Soil	3	0.34	0.683
PH x Str	1	0.00	0.987
PH x Ri	1	0.20	0.349
PH x MM	1	0.14	0.435
Sp x Soil	12	6.45	0.007
Sp x Str	4	1.36	0.200
Sp x Ri	**4**	**2.69**	**0.021**
Sp x MM	**4**	**2.40**	**0.034**
PH x Sp x Soil	11	2.00	0.636
PH x Sp x Str	3	0.19	0.843
PH x Sp x Ri	4	0.83	0.455
PH x Sp x MM	4	0.98	0.365
Residuals	184	39.19	



879

880 **Figure S1**. Mean root colonization by AMF of individual plants of the five plant species used
881 in Experiment 1 (see Table 1 in main text for species codes). Different shading indicates
882 different soil-history treatments. The non-AMF inoculated controls are not shown as no
883 colonization by AMF was detected. Error bars are standard errors of means.

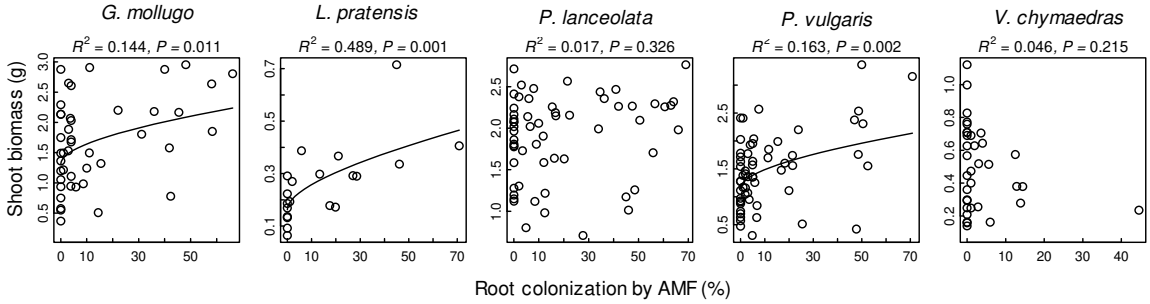
884



885
886 **Figure S2.** Total aboveground biomass, leaf dry matter content (LDMC) and leaf damage of
887 each of the five plant species in Experiment 1. Note there were no significant three-way inter-
888 actions of plant history by species by AMF-community history for these plant responses (see

889 Table 2). Plant history is indicated by different shading and AMF-community history is la-
890 beled below for each plant-history pair. Error bars are standard errors of the model estimates.
891 Plant species and trait abbreviations are defined in Table 1.

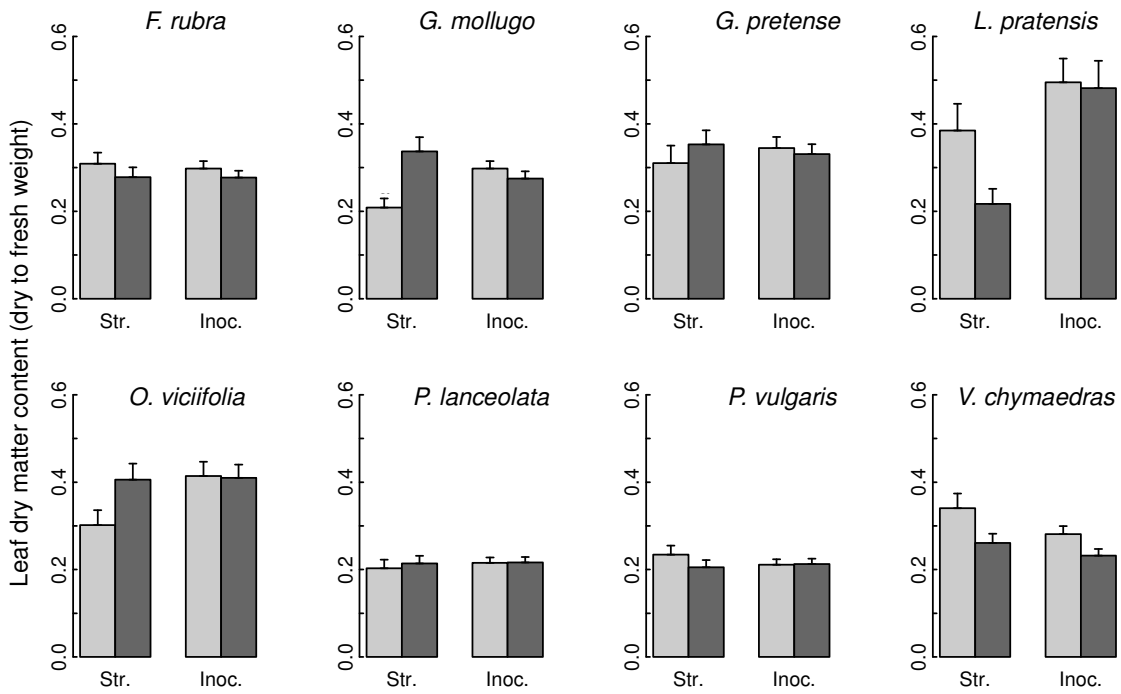
892



893

894 **Figure S3.** Scatter plots for the relationship between AMF root colonization and total
895 aboveground biomass for each of the five plant species in Experiment 1. Regression curves
896 are shown where relationships were significant (relationships were fit using y ~
897 log(colonization)). Fit statistics (R^2) and significances (P) are indicated above each panel for
898 each species.

899



900

901 **Figure S4**. Means and standard errors for the LDMC for each plant species inoculated with
902 either the sterile control inoculum (Str.) or with a non-AMF inoculation (Inoc.) illustrating
903 the three-way plant history x species x control vs living inoculum interaction in Experiment 2
904 (see Table 3 and Fig. 2 in main text). Light bars indicate plants selected in monoculture
905 history and dark bars plants selected in mixture. Error bars are standard errors of the
906 estimated means.

907

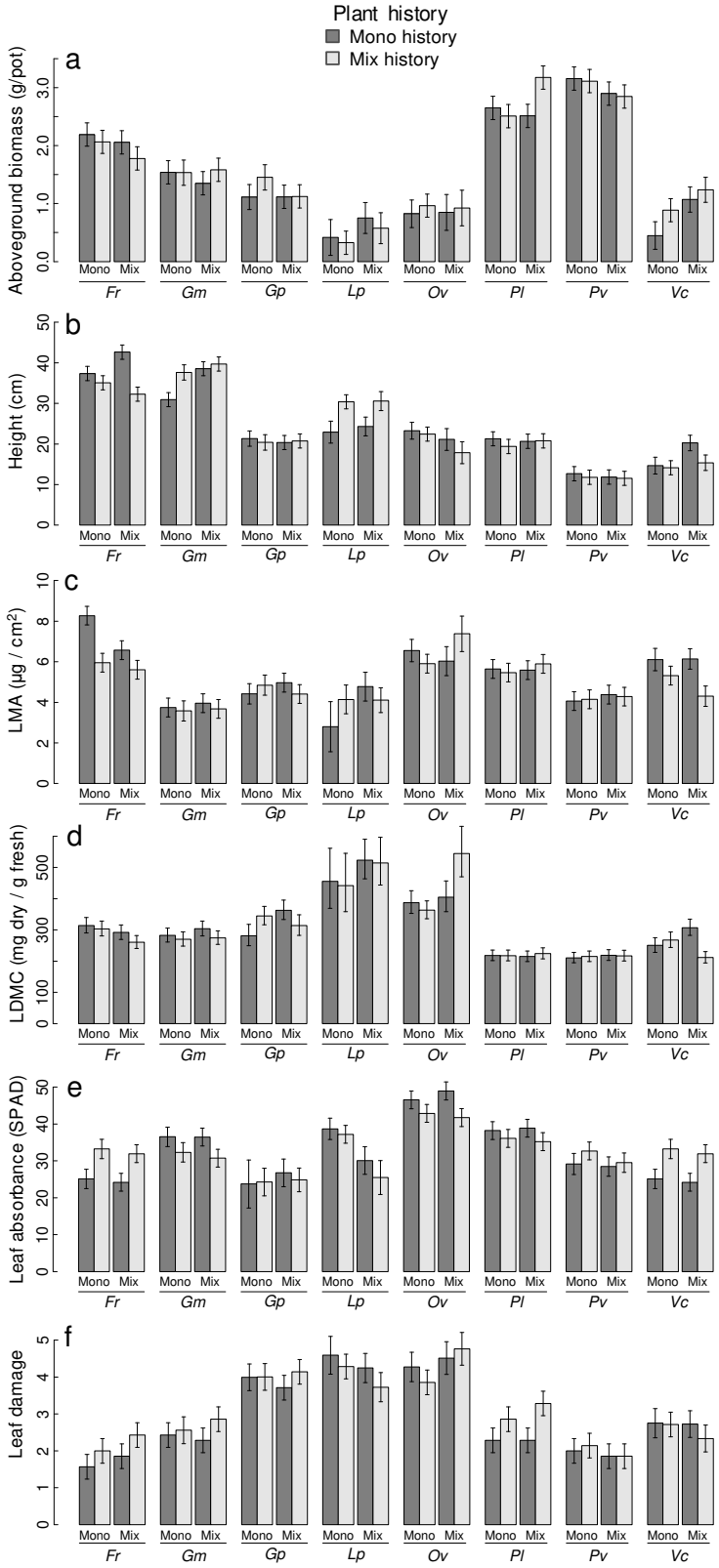


Figure S5. Mean of (a) aboveground biomass, (b) plant height, (c) leaf mass per area (LMA), (d) leaf dry matter content (LDMC), (e) leaf absorbance and (f) leaf damage of each of the

911 eight plant species in Experiment 2. Note there were no significant three-way interactions of
912 plant history by species by non-AMF community history for these plant responses (see Table
913 3). Plant history is indicated by different shading and non-AMF community history is labeled
914 below for each plant-history pair. Error bars are standard errors of the model estimates. Plant
915 species and trait abbreviations are defined in Table 1.
916